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EFFECT OF ENERGY SOURCE ON NUTRIENTS UTILIZATION AND RUMEN MICROBIAL ACTIVITY OF OSSIMI SHEEP

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ABSTRACT: *This experiment was carried out in order to study the mode of action of the dietary energy source on (digestibility, N-balance and rumen activity). Three Ossimi rams aged 3 years were used in a 3 × 3 Latin Square design. The first treatment was the control ration (clover hay + concentrate feed mixture) without additive (R_0); the second was the control ration plus 46g dry commercial fat (2.65% on DM basis, R_F,) while the third was the control ration plus 85g corn grain (4.88% on DM basis, R_C).*

Data reveal that dietary energy source significantly (P<0.01) increased intakes of DM, OM, EE and NFE. Fat-supplementation showed the highest values EE intake while corn-supplementation showed the highest intakes for DM, OM, CP and NFE. Water intake followed the same pattern being more for energy-supplemented ration than the control group. Addition of fat increased (P<0.01) the digestibility of DM, CP and EE. Corn-supplementation also increased the digestion coefficient of DM, OM, CP and NFE. Digestibility of CF was higher (P<0.01) for the control group than the energy-supplemented groups. Dietary fat and corn grain supplementation improved nitrogen balance through the improvement of N digestibility. Values of ruminal pH before feeding were 7.43, 7.44 and 7.39 for the treatment groups $\boldsymbol{R_0}$, $\boldsymbol{R_F}$ and $\boldsymbol{R_C}$, respectively. Differences were not significant. At 2-hr post feeding, pH declined with all treatment groups to reach the lowest values (being 6.55, 6.67 and 6.57 for the same respective groups). Significant (p<0.01) differences among groups were found at all times post feeding being lower for the control and $\boldsymbol{R_C}$ ration than $\boldsymbol{R_F}$. Rumen total VFA did not different among the dietary treatments at 2hrs (post-feeding); however, it was significantly (P<0.01) differed at 4 and 6h post feeding. Total VFA were 8.85, 8.44 and 9.03meq/dl of rumen liquor before feeding for $\boldsymbol{R_0}$, $\boldsymbol{R_F}$ and $\boldsymbol{R_C}$, respectively. In general, VFA increased in the treated-groups to reach its peak at 4-hrs post feeding and declined thereafter. The highest values were reported for the corn-treated group being 9.03, 17.23 and 12.60meq/dl; at 0, 4 and 6h post-feeding, respectively; the respective values were 8.44, 15.39 and 9.73meq/dl for $\boldsymbol{R_F}$. Before feeding, NH_3-N was 14.27, 13.06 and 12.45 mg/dl rumen liquor, differences were significant. Ammonia-N increased after feeding to reach the highest values for all dietary treatments at 2-hrs being 22.79, 21.15 and 20.60 mg/dl rumen liquor; and it decreased thereafter. Concentration of NH_3-N was significantly higher (P<0.01) for the control group than both treated groups.

Key words: *Dietary energy source, digestibility, microbial activity, Ossimi sheep.*

INTRODUCTION

The addition of fat to the diets of ruminant animals can help in covering the requirements of energy for high productive performance without causing metabolic disorders that often associated with large intakes of grain (Simas *et al.,* 1998). To increase energy density without the rumen acidosis and depression in milk fat due to the use of high starch and low fiber diets, attention has been directed to the inclusion of fats and feed stuffs with high concentration of fat and oils in ruminant rations (Choi and Palmquist, 1999; Chan *et al.,* 1997; Simas *et al.,* 1997 and 1998; Zervas *et al*., 1998; Casals *et al*., 1999; Kowalski *et al*., 1999; Offer *et al*., 1999).

The published reports contain lots of conflictions about the effect of dietary fat on animal performance, digestion kinetics, rumen fermentation, blood constituents and carcass traits. Some reports indicate a negative effect on feed intake (Bendary *et al.*, 1994; Lough *et al.,* 1994; Talha, 1996). They reported that dry matter intake was lower with animals fed fat-supplemented than those on un-supplemented rations. Others (Zinn, 1988 and 1989; El-Bedawy, 1989) found that fat supplementation did not affect feed intake. Digestibility of DM, OM and ADF was reported to decrease with adding fat (Hill and West, 1991); however, others (Bayourthe *et al.,* 1993; El–Bedawy *et al.,* 1994; El–Bedawy *et al.,* 1996; Talha, 1996) found an increase in digestibility of almost all nutrients. Bendary *et al.* (1994) found that digestibility of OM and NFE were not affected but CP and EE were significantly increased in supplemented-fat ration compared with the control. White *et al.* (1992) reported that supplemental fat did not affect average daily gain and feed efficiency. Although, Krehbiel *et al.* (1995) found that average daily gain linearly decreased (P<0.01) as dietary fat increased. Zinn and Plascencia (1996) found that addition of fat to the high forage diet increased ADG.

The present research was conducted to study the effect of supplementing rations with different dietary energy sources i.e., corn and dry fat on nutrient digestibility, N-balance and rumen fermentation of Ossimi sheep.

MATERIALS AND METHODS

This experiment was carried out in the Farm of Animal Production Department of the Faculty of Agriculture, Menofiya University (Shebin El–Kom), in order to study the mode of action of the dietary energy source on (digestibility, N-balance and rumen activity). Three Ossimi rams aged 3 years with an average body weight of 49±2kg were used in this study. Animals were randomly allocated to three treatments in 3 × 3 Latin Square design. The first treatment was the control ration (clover hay + concentrate feed mixture) without additive (R_0); the second was the control ration plus 46g dry commercial fat (2.65% on DM basis, R_F,) while the third was the control ration plus 85g corn grain (4.88% on DM basis, R_C). Dietary energy of the second and third CFM was increased by 0.31 MCal/d (according to NRC, 2001). Lambs were fed at DM level of 3.5% body weight. The ratio of concentrate to roughage was 50: 50. The experimental concentrate mixtures are presented in Table (1). Rams were housed individually in metabolic crates (160m × 0.53m) as described by Maynard *et al* (1979) for separate collection of feces and urine. Rations and fresh water were presented once every day at 8.30h. Residuals were weighed daily every first week of the collection periods and subtracted from the offered amount to obtain the actual feed intake.

During the collection periods, feces were quantitatively collected at 8:00 a.m. before feeding; 10% from feces was withdrawn and dried to a constant weight in a forced air oven at 70ºC for 24h. Dry fecal samples were kept ground in plastic pocket for later analysis. Urine was collected daily and a 10% aliquot was composited and refrigerated till analysis for nitrogen.

Rumen fluid samples were taken during the last two days of the collection periods. The samples were collected using the stomach tube attached to a vacuum pump, before feeding and then at 2, 4 and 6h after feeding. Rumen *p*H was measured immediately after collection using a digital *p*H meter (Sophisticated microprocessor, *p*H meter). Rumen fluid was strained through four layer of cheesecloth into plastic containers and kept at -20^{o}C for later analysis. Half of the samples were acidified using concentrated ortho-phosphoric acid and 0.1N hydrochloric acid to determine the total volatile fatty acids (VFA). The second half of samples was alkaline using 0.1N NaOH to determine the concentration of rumen ammonia nitrogen.

The chemical analysis was carried out at the Laboratory of Nutrition, Department of Animal Production, Faculty of Agricultural, Minufiya University. The determination of dry matter (DM), organic matter (OM), crude protein (CP), crude fiber (CF), ether extract (EE), nitrogen-free extract (NFE) and ash in the feed, and feces were carried out according to AOAC (1990). Ammonia-nitrogen determination was carried out as soon as possible using the steam distillation method described by Ahmed (1976). Total volatile fatty acids (VFA) were measured according to AOAC (1990).

Table 1: Composition of the formulated experimental concentrate mixtures .

Ingredient	The experimental concentrate mixtures		
	Control ration R_0	Fat-supplemented R_F	Corn-supplemented R_C
	%		
Yellow corn	44.6	42.6	48.9
Soybean meal	11.9	11.4	11
Cottonseed meal	19.8	18.9	18.1
wheat bran	19.8	18.9	18.1
Protected fat	-	4.3	-
NaCl	1	1	1
Limestone	2	2	2
Mineral and Vitamin premix	0.3	0.3	0.3
Sodium bicarbonate	0.3	0.3	0.3
Di-calcium phosphate	0.3	0.3	0.3
Total	100	100	100

Data of the present study were analyzed using GLM procedure of the SPSS program, version 13 (SPSS, 1997). Data were analyzed using the following linear model: $Y_{ij} = \mu + A_i + e_{ij}$

where: Y_{ij} = the observation on the ij^{th} animal; μ = overall mean; A_i = the effect of the i^{th} dietary treatments (i = 1, 2, 3); e_{ij} = a random error assumed to be independently randomly distributed with zero mean and variance σ^2_e , i.e. NID (0, σ^2_e). All data measured as percentages were subjected to arc-sin transformation to approximate normal distribution before being analyzed.

RESULTS AND DISCUSSION

1. Nutrient intakes of Ossimi rams as affected by dietary energy source:

Data in Table (2) reveal that dietary energy source significantly (P<0.01) increased intakes of DM, OM, EE and NFE. Fat-supplementation showed the highest values for EE intake while corn-supplementation showed the highest intakes for DM, OM, CP and NFE. Water intake followed the same pattern being more (P<0.01) for energy-supplemented ration than the control group.

Canale *et al.* (1990) reported that energy supplementation increased intake of DM and net energy. However, different kinds of fat-supplementation did not affect feed intake (West and Hill, 1990; Jenkins and Jenny, 1992; Schuff and Clark, 1992; Wu *et al.,* 1993; Salfer *et al.,* 1995; Cervantes *et al*., 1996; Madison-Anderson *et al*., 1997; Weiss and Wyatt, 2003). Others (Simas *et al*., 1997; Patton, 2004) reported that DM intake tended to decrease when rations were supplemented with different fat sources. The conflict in the response of experimental animals in the above mentioned studies may have been due to the effect of fat sources used on the appetite. Mullins *et al*. (2010) found that DM Intake was increased by Holstein cows fed diets containing different levels of wet corn gluten feed.

Table 2: Nutrient intakes of Ossimi rams fed different energy sources.

Treatment*	Intake of						
	DM kg/d	OM kg/d	CP g/d	CF g/d	EE g/d	NFE g/d	Water l/d
R_0	1.52[c] ± 0.37	1.31[c] ± 0.32	217.79[b] ± 0.51	406.15 ± 1.27	49.36[c] ± 0.12	641.14[c] ± 1.33	5.23[b] ± 0.11
R_F	1.55[b] ± 0.26	1.34[b] ± 0.22	218.87[b] ± 0.35	407.22 ± 0.87	79.53[a] ± 0.08	636.04[b] ± 0.91	6.19[a] ± 0.10
R_C	1.59[a] ± 0.34	1.37[a] ± 0.29	220.58[a] ± 0.46	405.84 ± 1.13	52.22[b] ± 0.10	691.72[a] ± 1.19	6.11[a] ± 0.11
Sig.	0.01	0.01	0.01	NS	0.01	0.01	0.01

[(*)] R_0, R_F and R_C, are control ration without or supplemented with 47g/d fat or 85g/d corn grain, respectively.
[a,b] Means with different superscripts within each column for each parameter are different (P<0.01). NS, not significant

2. Nutrient digestibility by Ossimi rams as affected by dietary energy source:

Table (3) presents the nutrient digestibility by Ossimi rams as affected by dietary energy source. Addition of fat increased (P<0.01) the digestibility of DM, CP and EE. Corn-supplementation also increased the digestion coefficient of DM, OM, CP and NFE. Digestibility of CF was higher (P<0.01) for the control group than the energy-supplemented groups.

Holter *et al*. (1992) reported that digestibility of ether extract was higher with fat supplemented diets. However, digestibility of CP was not affected by fat supplementation. Andrae *et al*. (2000) used sixty crossbred beef steers and found that digestibility of DM, OM, starch, and GE was greater (P<0.05) for the high-oil diet than the control diet, but lipid digestibility did not differ among treatments. Baraghit et al. (2003) reported that digestibility of DM, OM and CP was higher for fat-supplemented-followed by oil-supplemented-ration than the control one. El-Bedawy *et al*. (2004) found that 4% protected fat did not affect DM digestibility but increasing fat level to 8% decreased (P<0.05) DM digestibility by about 4 units. Digestibility of OM and CP was improved by incorporation of 4% dietary fat.

3. Nitrogen balance by Ossimi rams as affected by dietary energy source:

Results of nitrogen balance (Table 4) revealed that sheep fed all the experimental rations had almost similar nitrogen intake (NI) being 34.85, 35.02 and 35.29g/d. This could be attributed to that all the experimental ration were iso-nitrogenous.

The differences of fecal nitrogen (FN) between the control and the other groups were significant (P<0.01). Sheep on R_F excreted significantly (P<0.01) less N in the feces (10.03g) than the other groups (11.24 and 10.89g for R_0 and R_C, respectively). This resulted in better crude protein digestibility. Animals released almost equal amounts of N in the urine being 18.84, 18.25 and 18,21g for R_0, R_F and R_C groups, respectively. The NB values were significantly (P<0.01) more in R_F (6.74g) and R_C (6.19g) than the control group (4.76g).

It is obvious that dietary fat and corn grain supplementation improved nitrogen balance through the improvement of N digestibility. El-Bedawy *et al*. (2004) reported that feeding 4% fat containing rations improved (P<0.05) nitrogen retention by 15% in comparison with control or 8% fat rations.

Table 3: Nutrient digestibility (%) by Ossimi rams fed different energy sources.

Treatment*	DM	OM	CP	EE	NFE	CF
R_0	67.83[b] ± 0.24	67.49[b] ± 0.21	67.82[c] ± 0.21	69.46[b] ± 0.30	72.97[b] ± 0.23	58.50[a] ± 0.30
R_F	69.06[a] ± 0.38	68.06a[b] ± 0.35	71.40[a] ± 0.46	81.85[a] ± 0.30	72.51[b] ± 0.32	56.60[b] ± 0.47
R_C	68.96[a] ± 0.20	68.67[a] ± 0.26	69.17[b] ± 0.34	69.88[b] ± 0.47	74.83[a] ± 0.20	57.87[a] ± 0.36
Sig.	0.01	0.05	0.01	0.01	0.01	0.01

[(*)] R_0, R_F and R_C, are control ration without or supplemented with 47g/d fat or 85g/d corn grain, respectively.
[a,b] Means with different superscripts within each column for each parameter are different (P<0.01). NS, not significant.

Table 4: Nitrogen balance (g/d) by rams fed different energy sources.

Treatment*	NI	FN	UN	NB
R_0	34.85[b] ± 0.08	11.24[a] ± 0.08	18.84[a] ± 0.13	4.76[b] ± 0.13
R_F	35.02[b] ± 0.06	10.03[b] ± 0.16	18.25[b] ± 0.16	6.74[a] ± 0.25
R_C	35.29[a] ± 0.07	10.89[a] ± 0.11	18.21[b] ± 0.17	6.19[a] ± 0.21
Sig.	0.01	0.01	0.01	0.01

[(*)] R_0, R_F and R_C, are control ration without or supplemented with 47g/d fat or 85g/d corn grain, respectively.
[a,b] Means with different superscripts within each column for each parameter are different (P<0.01). NS, not significant.

4. Rumen activity by Ossimi rams as affected by dietary energy source:

Data of sheep rumen fermentation (*p*H, VFA and NH_3-N) as affected by dietary energy source are illustrated in Figures (1-3).

Values of *p*H before feeding were 7.43, 7.44 and 7.39 for the treatment groups R_0, R_F and R_C, respectively. Differences were not significant. At 2-hr post feeding, *p*H declined with all treatment groups to reach the lowest values (being 6.55, 6.67 and 6.57 for the same respective groups). Significant (p<0.01) differences between groups were found at all times post feeding being lower for the control and R_C ration than R_F. The lower *p*H was due to the higher fermentation ability of corn grain. Logically, *p*H values took the opposite trend of the total VFA.

Steele *et al.* (2012) reported that rumen *p*H for cattle fed high grain diet displayed lower rumen *p*H compared with cattle receiving the control diet. Zened *et al*. (2013) showed that rumen *p*H 5-h post feeding decreased from 6.4 to 5.7 when starch was added to the diet. Many studies

(Ohajuruka *et al*., 1991; Knapp and Grummer, 1991; Schauff and Clark. (1992) Madison-Anderson *et al*., 1997; Abdelqader and Oba, 2012) reported that fat supplementation did not affect rumen *p*H.

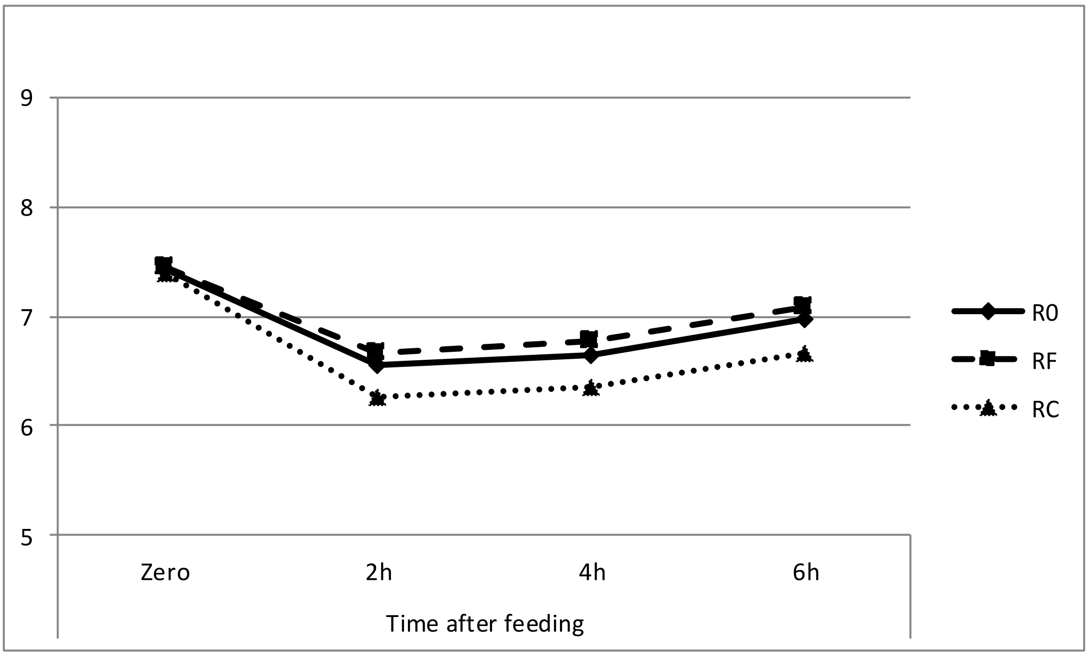

Fig. 1: Rumen pH rams fed different energy sources.

Rumen total VFA (Fig 2) was not different among the dietary treatments at 2hrs (post-feeding); however, it was significantly (P<0.05) different at 0 time (before feeding) and (P<0.01) at 4 and 6hrs post feeding. In this respect Values were higher for the control group than the treated ones. Total VFA was 8.85, 8.44 and 9.03meq/dl of rumen liquor before feeding for R_0, R_F and R_C, respectively. In general, VFA increased in the treated-groups to reach its peak at 4-hrs post feeding and decline thereafter. The higher values were reported for control and the corn-treated group. The values of corn-treated group being 9.03, 17.23 and 12.60meq/dl; at 0, 4 and 6h post-feeding, respectively; the respective values were 8.44, 15.39 and 9.73meq/dl for R_F.

The lowest VFA values reported for the dry fat-treated ration may have been due to a lower microbial activity in the rumen of sheep fed this ration than those received the control or corn-supplemented rations, perhaps because it contained less fermentable carbohydrate; it is worthy to mention that the control ration also contained more corn than the fat-supplemented ration (Table 1).

The published articles have some conflicts regarding the effect of dietary fat on rumen VFA concentration. Differences may have been due to different factors such as the form and/or level of fat used, the level of the dietary energy, the other ingredients in the diet, feeding frequency etc. Schauff and Clark (1992) reported that total VFA's concentrations were decreased when Ca-soaps was added to the diet. Madison-Anderson *et al*. (1997) reported that rumen total VFA's concentrations were greater for cows fed the control diet than those fed fat supplemented diet. Zened *et al*. (2013) reported that total VFA were significantly higher in the rumen of cows receiving high starch (HS) and high starch + sunflower oil (HS+OL) diets than in those of control CON and OL diet fed cows being 133 vs. 98 mM (on average), respectively. Sun and Oba

(2014) reported that cows fed the dried distillers grains with soluble (DDGS) diet tended to have higher total VFA concentration in rumen fluid compared with cows fed the CON diet (107 vs. 116 mM; P = 0.06).

Data of ammonia nitrogen concentration in the rumen of sheep as affected by the dietary energy source are presented in Fig (3). Before feeding, NH_3-N was 14.27, 13.06 and 12.45 mg/dl rumen liquor, differences were significant. Ammonia-N increased after feeding to reach the highest values for all dietary treatments at 2-hrs being 22.79, 21.15 and 20.60 mg/dl rumen liquor; ammonia-N decreased thereafter. Concentration of NH_3-N was significantly higher (P<0.01) for the control group than the treated groups. Demeterova *et al*. (2002) investigated the influence of Ca-soaps on rumen fermentation. They reported that NH_3-N decreased for cows fed diets contained Megalac.

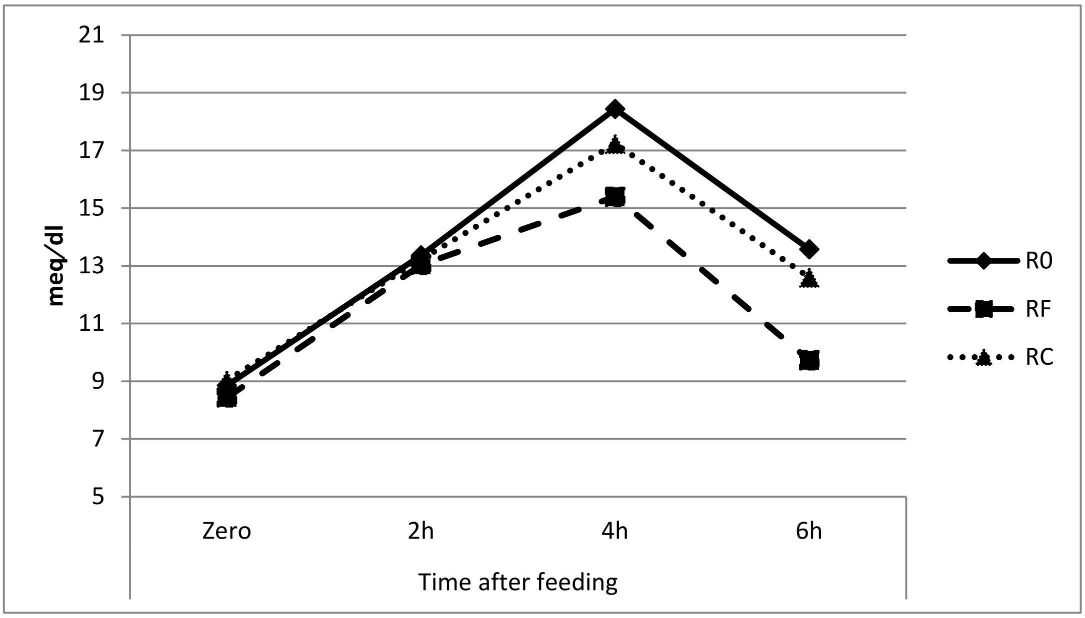

Fig. 2: Rumen totl VFA concentration (meq/dl) by rams fed different energy sources.

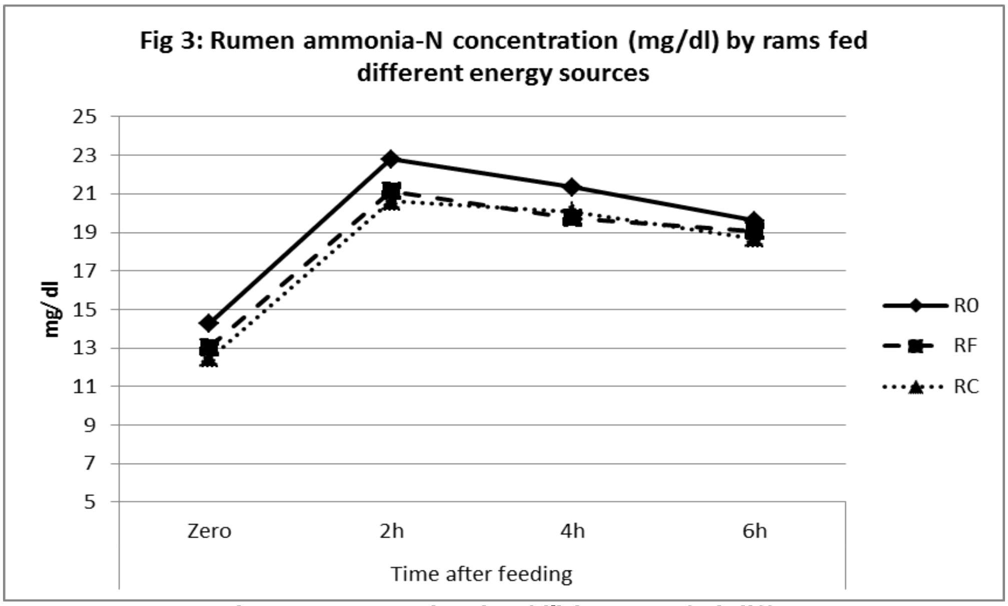

Fig. 3: Rumen ammonia-N concentration (mg/dl) by rams fed different energy sources.

From the present study it could be recommended that sheep diets should be supplemented with energy source, either dry fat or corn in order to improve intake, digestibility, N balance as well as rumen fermentation.

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تأثير مصادر طاقة الغذاء على الاستفادة من الغذاء والنشاط الميكروبي بكرش الأغنام الأوسيمي

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الملخص العربى

أجريت التجربة بهدف دراسة تأثير إضافة مصادر طاقة مختلفة بعلائق الأغنام الأوسيمي على كفاءة الهضم والإتزان النيتروحيني وتخمرات الكرش. أستخدم ثلاثة كباش عمر 3 سنوات في تصميم تجريبي 3×3 مربع لاتيني – المعاملة الأولى عليقة المقارنة مكونة من دريس البرسيم ومخلوط مركزات بينما في الثانية إضيف 46جم دهن جاف إلى المخلوط المركز وفي الثالثة أزيدت كمية الذرة في مخلوط المركزات بمقدار 85جم. أشارت النتائج إلى أن مصدر الطاقة الغذائية كان له تأثيرا إيجابيا على كميات الغذاء المستهلك وكان التأثير الأكبر لدهن الغذاء على المأكول منه بينما أدت إضافة الذرة إلى زيادة المأكول من المادة الجافة والعضوية والبروتين والكربوهيدرات. تحسنت معاملات الهضم نتيجة إضافة مصادر الطاقة عدا الألياف الخام فقد كانت معاملات هضمها أكبر في العليقة المقارنة. أدت إضافة الطاقة من مصادرها المختلفة إلى تحسن الاتزان النيتروجيني. تحسنت تخمرات الكرش في المعاملة المضاف لها الذرة عن إضافة الدهن. انخفضت قيمة الحموضة بالكرش بعد التغذية لتصل إلى الحد الأدنى عند ساعتين ثم بدأت في الارتفاع ثانية. التغذية على الذرة كمصدر إضافي للطاقة أدى إلى ارتفاع تركيز الأحماض الدهنية الطيارة عن المعاملة بالدهن – وقد تبعت حموضة الكرش الاتجاه المعاكس بينما لم تتأثر تركيزات أمونيا الكرش بنفس الدرجة.