

EFFECT OF PROTEIN LEVELS IN RATION ON GROWTH PERFORMANCE AND RUMEN ACTIVITY IN BUFFALO CALVES .

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ABSTRACT: Fifteen buffalo calves of 251.53-Kg average live body weight were divided into three experimental groups each of 5 calves. The 3 groups received 3 diets containing 3 levels of CP, 12,14 and 16%. Performance, digestibility and rumen liquor chemistry were assessed during the trial, which lasted for an average of 120 days to reach a certain final body weight. The results revealed that daily body gain (DBG) increased as the level of protein increased and although the feed conversion was improved yet the change was not significant. Days to final body weight were significantly ($P>0.05$) less at 16%(112 days) and 14%(118.4) protein level respectively than at 12%(130.4). Digestibility of organic matter was significantly ($P\leq 0.05$) increased at 16%CP (68.85%), likewise was digestibility of crude protein which increased with increasing the level of crude protein in the diet being 62.8,64.4 and 68.2% at 12,14 and 16%CP respectively. Similarly was the trend with Ether extract, crude fibers and N.F.E. Total volatile fatty acids and N- ammonia increased significantly with the level of protein, the former lead to decreased rumen PH. Total count of bacteria in liquor decreased significantly ($P\leq 0.05$) while total count of protozoa increased ($P\leq 0.05$) as the level of protein in the diet increased.

INTRODUCTION

In many countries around the world, low quality roughage is the only feed available to grazing animals most of the year. Ruminant animals can utilize large quantities of low quality roughage more efficiently and transform it to animal products. Effect of the low protein content on the intake or

utilization of low quality roughage has been recognized (Hennessy et, al., 1983; Willms et, al.and 1991; El -Taweel, 2000). Supplementary protein has been shown to improve live body gain (Titi et, al., 2000) and digestibility (Solaiman et, al., 1990; Willms et, al., 1991 and El-Taweel, 2000). Ørskov, (1977) suggested that for rapid growth rates in ruminants, microbial protein production in the rumen cannot meet the needs of the host animal. High levels of protein feeding may be effective in promoting rapid live weight gains, but because of partial degradation in the rumen, such protein is likely to be inefficiently utilized .The objective of this study, therefore, was to investigate the effects of 3 protein levels on feed intake and efficiency, digestibility of nutrients, rumen liquor chemistry pH, total volatile fatty acid, ammonia-N, total count of bacteria and protozoa, and economic efficiency.

MATERIAL AND METHODS

Fifteen male buffalo calves (average initial weight of 251.53 kg), were divided into three groups, each group was fed on one of the protein levels tested (12.14 and 16%). The three diets formulated and used in the experiment are shown in (Table, 1). The animals were fed *ad-libitum* and the diets were offered individually twice daily at (8.00 and 1600 hrs.), while, water was offered 3 times / day. Three digestibility trials were carried out using five male buffalo calves representing each treatment. Animals were individually placed in metabolism cages for 10 days for adaptation and 5 days for feces collection. Feces were collected daily and a 5% (w/w) sub samples were taken and stored in plastic bags at 4°C and left to be composted over the collection period.

Table (1): Ingredients forming the rations at 3 levels of CP.

Ingredient %	% Dietary CP		
	12	14	16
Soybean meal	12	18	23
Rice bran	23	17	12
Wheat bran	20	20	20
Corn stalk	30	30	30
Corn cobs	5	5	5
Molasses	7	7	7
Calcium carbonate	2	2	2
Sodium chloride	1	1	1
TDN (calculated)	55.32	56.64	57.74

Approximately 400g of fecal composted samples were dried at 50°C and ground to pass through 1-mm screens for chemical analysis. Diet samples were collected during each trial. Crude fiber and ether extract in diets and in feces were determined according to (A.O.A.C, 1990). Crude protein was determined by the Kjeldahl method (A.O.A.C, 1990). Ruminant liquor samples were collected pre feeding, at 3 and 6h post feeding in the last day of digestibility trials by stomach tube from five calves of each group for measurement of the P^H, ammonia-N, total volatile fatty acids and total count of bacteria and protozoa. The P^H value was measured immediately after collection by using pH meter. TVFA were determined by the distillation method according to Warner (1964). Some of rumen liquor was used for ammonia determination directly as described by Abou-Akkada and Osman (1967). Total count of bacteria was determined by using trypton glucose extract agar medium as recommended by the American public Health Association (1978), while, total count of Protozoa was done as described by El-Saifi (1969).

The analysis of variance procedure was used to test the effect of treatments. Least significant difference method was used for comparison among means. ANOVA of SAS (S A Inc. (1988), was used for statistical analysis.

RESULTS AND DISCUSSION

Performance of buffalo calves as affected by different protein levels are shown in table, 2. Buffalo calves fed on the ration containing 16% CP had higher (P<0.01) average daily gain than those fed on the other rations, while, calves fed on the ration containing 12% CP had significantly lower (P<0.01) daily gain than those fed 14 or 16% C.P. Calves fed on the 12% CP ration consumed less feed than the other 2 treatment groups. Depression of feed intake was expected in calves fed 12%CP; these small differences may explain partially the differences in daily body gain. Increase in daily gain with increasing protein level may be due to increased flow of non-degradable true protein and microbial protein to the abomasum. The present results are supported by those obtained by Bergen et al. (1982) and Firkins et al., 1986) who proposed that the intake; digesta passage and out flow under *ad. Lib*

feeding condition should increase total microbial cell protein production and (or) flow to the lower gut. The present result also conforms the recommendations of Titi et al., (2000) and El- Taweel, (2000) who reported that daily gain increased with the increase of protein level in diets of lambs, goats, Holstein - Friesian male calves and buffalo male calves.

Table (2): Effect of protein levels on growth performance of buffalo calves.

	% Dietary CP		
	12	14	16
Initial weight Kg	251.40 ± 0.75 ^A	252.00 ± 0.89 ^A	251.20 ± 2.44 ^A
Final weight Kg	352.00 ± 1.58	358.60 ± 2.08	369.00 ± 2.44
Days to final weight	130.40 ± 5.10 ^A	118.4 ± 3.96 ^B	112.00 ± 0.00 ^B
Body gain (Kg/Animal)	100.6 ± 0.98	106.6 ± 1.72	117.8 ± 2.45
Daily body gain (Kg/Anim./day)	0.77 ± 0.03 ^C	0.91 ± 0.03 ^B	1.05 ± 0.02 ^A
Feed consumption (Kg. DM. /Anim.)	723.60 ± 28.69	756.80 ± 21.35	791.60 ± 3.59
Feed conversion (Kg. DM.feed/Kg gain)	7.19 ± 0.31 ^A	7.11 ± 0.25 ^A	6.73 ± 0.15 ^A

(A, B) Means with the same superscript are not significantly different at level of (P < 0.05).

Feed conversions (Table, 2) as affected by level of protein in the rations were 7.19, 7.11 and 6.73 kg DM feed/kg gain for rations containing 12, 14 and 16% CP, respectively. The improvement in feed conversion of these rations may be due to the increase in feed consumption, accordingly reduced degradability of feed protein at higher CP contents. A direct evidence shows that at higher feeding rate, feed protein degradability was reduced (Tamminga, 1981). The effect of reduction of protein degradability would lead to increase in the amount of CP needed in a ration to meet the rumen degradable protein needs of rumen microorganisms. Similar results were obtained by El-Serafy et al., (1979), who found that feed efficiency (g gain / intake) on DM basis was improved with increasing concentrate level. Also the same trend was observed by Petit and Flipot, (1992), Rossi-JE et al., (1999) and El-Taweel, (2000). Improvement of daily body gain and feed conversion with increasing protein level was reflected on the elapsed period (days to final body weight). This period was significantly (P<0.01) shorter at higher protein level (14% CP and 16% CP).

Apparent digestibility (AD) values for the three rations are listed in table, (3) and figure (1). OM, CP, EE, CF and N-Free extract were significantly ($P<0.05$) higher for the ration containing 16% CP than for the other rations. Differences in A.D. between rations containing 12 and 14 % CP in CP, EE, CF and N-Free extract were significant however differences between these rations in OM digestibility was not significant. These results showed increased apparent digestion coefficients with increasing CP level in the ration. There results may due to increased soybean meal (used in this experiment) with higher digestibility than other ingredients included in the diet (Seoane and Moore, 1969) or increased microbial activity with increasing concentrate level in the diet (El-Taweel, 2000).

Table(3):Effect of protein levels on apparent digestibility (A.D.) of experimental rations.

Digestibility %	% Dietary CP		
	12	14	16
OM	63.96 ± 0.23 ^B	65.41 ± 0.52 ^B	68.85 ± 0.76 ^A
CP	62.82 ± 0.31 ^C	64.42 ± 0.13 ^B	68.24 ± 0.55 ^A
EE	55.86 ± 0.21 ^C	58.55 ± 0.41 ^B	60.58 ± 0.53 ^A
CF	60.86 ± 0.16 ^C	62.63 ± 0.38 ^B	65.77 ± 0.11 ^A
NFE	64.77 ± 0.33 ^C	66.49 ± 0.45 ^B	69.73 ± 0.49 ^A

(A, B) Means with the same letter are not significantly different at level of ($P<0.05$).

Two factors concerning rumen metabolism are likely to be important in affecting the quality of feed and ration digestibility:

- 1- The effect of feeding on retention time of feed in the rumen.
- 2- Implication for microbial growth (Oldham, 1984).

Willms et al, (1991) reported that apparent total tract of DM, OM, NDF and ADF digestibility all increased linearly ($P<0.01$) with increasing CP level, for ram lambs fed on 6,8,10,12,14 or 16% CP of the DM contents. They found also that nitrogen retention expressed as a percentage of N intake and as g. /day were greater in lambs fed 14% CP. Significant increase in OM, CP, EE, and N-free extract digestion coefficients were noticed accordingly

with increasing level of soybean meal in the rations, for buffalo calves fed with out or with 0.5kg soybean meal/day/animal (El-Taweel. 2000). Table (4) and Figure (2) show the changes in pH, ammonia-N and TVFA in rumen liquor at different protein levels in the rations and sampling time. Regardless of protein levels, pH value was significantly ($P<0.01$) higher before feeding and was decreased at 3hr post-feeding, then it was significantly ($P<0.05$) re-increased, while, TVFA and ammonia-N were significantly ($P<0.05$) and linearly increased with increasing sampling time even 3hr after feeding and then decreased significantly, but remained significantly higher compared to their value before feeding.

Table (4): Effect of protein level on pH, ammonia-N and TVFA concentration in rumen liquor.

	*	Dietary CP%			Overall mean
		12	14	16	
pH.	0	7.33 ± 0.07	7.05 ± 0.05	6.85 ± 0.03	7.07 ± 0.13 ^A
	3	6.91 ± 0.06	6.72 ± 0.08	6.37 ± 0.03	6.66 ± 0.15 ^C
	6	7.05 ± 0.05	6.85 ± 0.07	6.56 ± 0.10	6.82 ± 0.14 ^B
Overall mean		7.09 ± 0.12 ^a	6.87 ± 0.09 ^b	6.59 ± 0.16 ^C	
Ammonia-N (m.eq./100ml.)	0	14.38 ± 0.18	17.46 ± 0.15	19.44 ± 0.17	17.09 ± 1.47 ^C
	3	23.16 ± 0.19	26.33 ± 0.10	28.94 ± 0.37	26.14 ± 1.67 ^A
	6	18.36 ± 0.18	22.99 ± 0.26	24.91 ± 0.34	22.08 ± 1.95 ^B
Overall mean		18.63 ± 2.54 ^C	22.26 ± 2.59 ^b	24.43 ± 2.75 ^a	
TVFA (m.eq./100ml.)	0	6.63 ± 0.08	7.89 ± 0.06	8.79 ± 0.12	7.77 ± 0.63 ^C
	3	8.08 ± 0.08	9.00 ± 0.29	9.89 ± 0.03	8.99 ± 0.52 ^A
	6	7.77 ± 0.13	8.56 ± 0.28	9.32 ± 0.04	8.55 ± 0.45 ^B
Overall mean		7.49 ± 0.44 ^C	8.48 ± 0.32 ^b	9.33 ± 0.31 ^a	

Means in rows or columns with the same letter (a,b,...)or(A,B,...)are not significantly different at level of ($P<0.05$). Interaction between treatment and time of sampling are not significant. (*) Time of sampling past feeding.

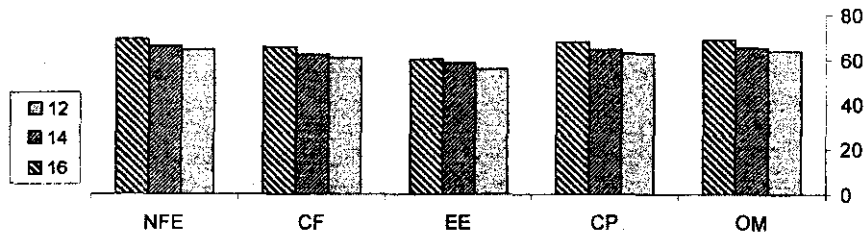


Fig. (1): Effect of protein levels on digestibility.

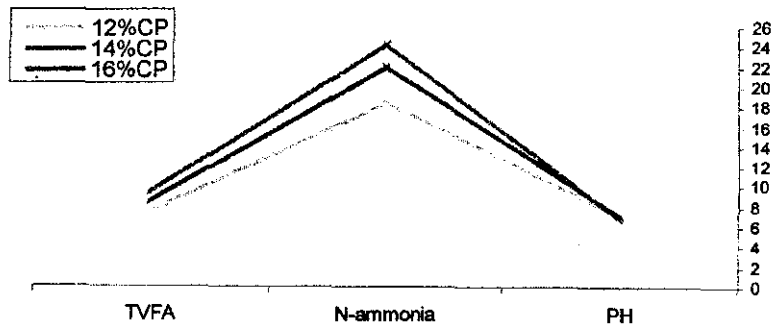


Fig. (2): Effect of protein levels on pH, Ammonia-N and TVFA concentration in rumen liquor.

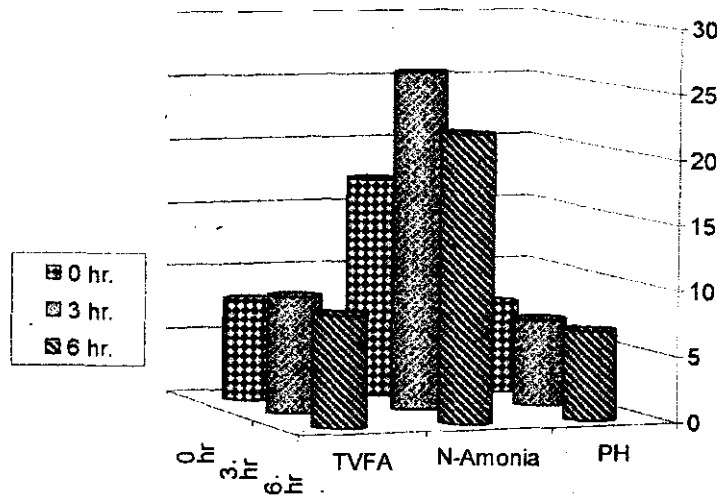


Fig. (3): Effect of Sampling time on pH, Ammonia-N and TVFA values in rumen liquor.

These results may indicate increased fermentation and microbial action on feed with time elapsing after morning feeding. Nadan et al., (1979), noticed that with sheep the higher production of VFA and ammonia-N achieved at 4hr. after feeding perhaps because most of the biochemical changes in the rumen might have been completed. Similar results have been obtained by El-Taweel, (2000) who found that pH was decreased and TVFA or ammonia-N concentration were increased significantly even at 3hr, then increased and decreased significantly. Irrespective of sampling time, all differences among means in TVFA and ammonia-N were significant ($P<0.05$). Calves fed on 12%CP diet recorded the less TVFA and ammonia-N in the rumen liquor, while, the diet which contained 16%CP resulted in more TVFA and N-ammonia followed by the diet which contained 12% CP. Analysis of variance indicated that pH value had an opposite trend to that of TVFA concentration. Ration containing 12% CP recorded higher value of pH of in rumen liquor. pH value was significantly ($P<0.01$) decreased with increasing protein level in the ration. Fouad, (1991) reported that rumen pH value was decreased with increasing the VFAS concentration. Results obtained in the present study indicated that CF digestibility was increased with increasing protein level in the ration.

El-Taweel, (2000) found a linear positive correlation between TVFA and ammonia-N concentration in rumen liquor and crude fiber digestibility. Abd-El-Hafez, (1983) reported that TVFA concentration in Ballady goats rumen liquor increased with increasing concentrate level in the ration. Similar results also were obtained by Delcurto et al, (1990) Willms et al., (1991) and Yan et al., (1996), they reported that ammonia-N concentration in rumen liquor was increased with addition of soybean meal to barley diet.

Differences in total count of bacteria and protozoa (Table,5) among diets containing 12,14 and 16% CP were significant ($P<0.05$), except, the difference in total count of protozoa between 14 and 16% CP which was not significant. The ration containing 16% CP recorded significantly ($P<0.01$) the lowest value of total count of bacteria compared to other diets, while, the ration containing 12% CP recorded significantly ($P<0.05$) the lowest value of total count of protozoa compared to other diets. Regardless of protein level

in the ration, total count of protozoa and bacteria in rumen liquor increased and decreased significantly ($p < 0.05$) with elapsing time after feeding. A non-significant decrease in bacterial count from 3 hr to 6hr was noticed. These results indicated that total count of protozoa increased with time after feeding or protein level, which may be due to increased fermentation after feeding or due to increasing protein level in the ration. Helali, (1986) noticed that activity and count of protozoa increased with increased fermentation in the rumen. Increase of total count of protozoa was accompanied with a decrease in total count of bacteria under effect of the increase in protein level or the time after feeding which may suggest that the protozoa was fed on bacteria, Helali, (1986) noticed that protozoa obtained nitrogen by feeding on bacteria. He also mentioned that count of bacteria was decreased with increased count of protozoa. The present results confirm with that obtained by Kandil, (1998) on sheep and El-Taweel, (2000) on buffalo calves, who reported that the count of bacteria was decreased and protozoa increased with increase in concentrate level in the ration or with time after feeding.

Table(5): Effect of protein levels on Total count of bacteria and protozoa in rumen liquor.

	*	% Dietary CP			Overall mean
		12	14	16	
Total count bacteria ($\times 10^9/\text{ml}$)	0	1.11 \pm 0.03	1.09 \pm 0.03	1.06 \pm 0.08	1.08 \pm 0.01 ^A
	3	1.09 \pm 0.03	1.07 \pm 0.05	1.06 \pm 0.01	1.07 \pm 0.08 ^B
	6	1.08 \pm 0.05	1.06 \pm 0.03	1.05 \pm 0.05	1.06 \pm 0.08 ^B
Overall mean		1.09 \pm 0.08 ^a	1.07 \pm 0.08 ^b	1.05 \pm 0.03 ^c	
Total count protozoa ($\times 10^3/\text{ml}$)	0	1.02 \pm 0.03	1.04 \pm 0.03	1.04 \pm 0.05	1.03 \pm 0.01 ^C
	3	1.04 \pm 0.03	1.05 \pm 0.03	1.05 \pm 0.05	1.04 \pm 0.03 ^B
	6	1.04 \pm 0.03	1.05 \pm 0.03	1.06 \pm 0.03	1.05 \pm 0.05 ^A
Overall mean		1.03 \pm 0.06 ^b	1.04 \pm 0.03 ^a	1.05 \pm 0.05 ^a	

Means in rows or columns with the same letter (a,b,...) or (A,B,...) are not significantly different at level of ($P < 0.05$). Interactions between treatment and time of sampling were not significant. (*) Time of sampling post feeding.

Table (6): Economic evaluation of experimental ration.

	% Dietary CP		
	12	14	16
Feed consumption (Kg)	723.60 ± 28.69	756.80 ± 21.35	791.60 ± 3.59
Price of Kg feed (L.E)	0.37	0.40	0.43
Feed costs (L.E)	267.73 ± 10.66	302.64 ± 8.62	340.39 ± 1.55
Body gain (Kg)	100.60 ± 0.98	106.60 ± 1.72	117.8 ± 2.45
Feed costs /Kg gain (L.E)	2.66 ± 0.15 ^A	2.84 ± 0.10 ^A	2.89 ± 0.06 ^A
Market Price of Kg body gain (L.E)	7.00	7.00	7.00
Market Price of body gain (L.E)	704.20 ± 6.87	746.20 ± 12.07	824.60 ± 17.25
Net revenue (L.E)	436.47 ± 13.78 ^A	443.48 ± 16.32 ^A	484.21 ± 17.07 ^A
Economic feed efficiency(%)	164.93 ± 12.32 ^A	147.44 ± 8.89 ^A	142.26 ± 5.03 ^A

(A, B) Means with the same letter are not significantly different at level of (P<0.05).

Feed costs per kg gain and net revenue (Table, 6) were 2.66, 636.41; 2.84, 443.48 and 2.89 and 484.21 L.E for the rations containing 12,14 and 16% CP, respectively. Differences among means were but not significant. The economic efficiency of the feed obtained in this study decreased insignificantly with each increase in protein level fed from 12 to 14or16%, but the ration containing 16%CP recorded higher net revenue and shorter time to final body weight (Table,2) than the other two rations.

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تأثير مستويات مختلفة من البروتين في العليقة على أداء النمو ونشاط الكرش في العجول الجاموسى

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استخدم فى هذه الدراسة عدد ١٥ عجل جاموسى متوسط وزنها ٥٣.٢٥١ كجم موزعة على ثلاثة مجموعات فى كل منها ٥ عجول وتم تغذيتها على ثلاث مستويات من البروتين ١٢ ، ١٤ ، ١٦ % بروتين خام وكانت أهم النتائج كما يلى :

- ١- الزيادة اليومية فى الوزن كانت تزيد معنوياً بزيادة مستوى البروتين (٧٧.٠٠ و ٩١.٠٠ و ١.٠٥ كجم للمستويات ١٢ و ١٤ و ١٦% بروتين خام على التوالي).
- ٢- كان هناك تحسن فى الكفاءة التحويلية والغذاء المستهلك مع زيادة مستوى البروتين فى العليقة .
- ٣- زيادة مستوى البروتين فى العليقة أدى إلى زيادة معنوية فى معامل هضم البروتين من ٨٢.٦٢% عند مستوى ١٢% إلى ٤٢.٦٤% عند مستوى ١٤% ثم إلى ٢٤.٦٨% عند مستوى ١٦% بروتين خام وكذلك أدى إلى زيادة معنوية فى معامل الهضم لكل من الدهن الخام والألياف الخام والمستخلص الخالى من الأزوت.
- ٤- درجة حموضة الكرش كانت تقل معنوياً بزيادة مستوى البروتين فى العليقة وعلى العكس من ذلك كان تركيز الأمونيا والأحماض الدهنية الطيارة فى سائل الكرش يزيد معنوياً بزيادة مستوى البروتين .
- ٥- كانت الزيادة فى العدد الكلى للبكتيريا فى سائل الكرش معنوية مع زيادة مستوى البروتين فى العليقة أما العدد الكلى للبروتوزوا كان يزيد معنوياً حتى مستوى ١٤% بروتين خام ثم يستمر فى الزيادة ولكنها غير معنوية حتى مستوى ١٦% بروتين خام .
- ٦- على الرغم من أن تكاليف العليقة كانت تزيد بزيادة مستوى البروتين وكان صافى الربح يقل تبعاً لذلك مع عدم وجود اختلافات معنوية فى الكفاءة الاقتصادية إلا أن المدة اللازمة للوصول إلى الوزن النهائى كانت (١١٢ و ٤.١١٨ يوم) أقل معنوياً عند مستوى ١٦ و ١٤% على الترتيب.