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RUMEN FERMENTATIONS, RUMEN CILIATE PROTOZOA AND SOME BLOOD PARAMETERS IN EARLY WEANED LAMBS FED DIETS WITH DIFFERENT CONCENTRATE: ROUGHAGE RATIO.

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SUMMARY

Early rumen fermentations, identification and density of rumen ciliate protozoa and some blood parameters protozoa were studied in eighteen early weaned Barki male lambs, with an average live body weight 8 kg at 8 weeks age. Lambs were randomly allocated in three groups (6 lambs each) according to body weight. Lambs in the three groups were fed starter pelleted consists of different concentrate: roughage ratios (T1: 90:10, T2: 80:20 and T3: 70:30). The experiments were lasted for sixteen weeks.

The main results indicated that, ruminal pH values were slightly affected by treatments. As for ruminal total volatile fatty acids, ruminal ammonia nitrogen, Non protein nitrogen, ruminal total nitrogen and ruminal true protein nitrogen concentrations they were significantly ($P<0.01$) affected by treatments. It seems that during the whole period T3 had the highest values followed by T2 while the lowest values were for T1. It was interest to show that, there was a significant increase ($P<0.01$) in concentrations of those parameters with advanced age from 8 weeks age to 24 weeks age. Those parameters also increased at 3 hours after feeding more than before feeding then it gradually decreased by the time of sampling. Six genera with 16 species and 6 subspecies or formae of ruminal protozoa in lambs were identified [*Entodinum, Dasytrachia, Isotrchia, Diplodinum, Polyolastron and Ophryoscolox sps.*]. The values of ruminal protozoa count and ratio were increased gradually by age progressed from 8weeks to reach the maximum value at 24 weeks.

The three treatments had no significant effect on total serum proteins, albumin, globulin and glucose. The values were increased at 4 and 8hrs post-feeding compared to pre-feeding values. T2 had the lowest value of serum urea, while T1 and T3 were almost the same values. The values were high at zero time then it decreased ($P<0.01$) by progressed time of feeding. Lambs of diet T2 and T3 had significantly lowered ($P<0.01$) serum triglyceride than those of T1, the values were significantly ($P<0.01$) increased by progressed time of feeding, the highest value was at 8hrs post feeding,

***Keywords**: lambs, early weaned, rumen parameters, rumen ciliate protozoa and blood parameters*

INTRODUCTION

The practice of weaning lambs completely off milk at an early age, from which time they received only concentrates, hay and water, requires early development of the ruminant stomach. The proportion of concentrates to roughages in the diet of early weaned animals is very variable.

Lambs begin to consume solid feed between 2 and 4 weeks of age. The fermentation of this feed by ruminal microorganisms results in VFA production (Van Houtert, 1993). The factors that stimulate rumen metabolic development are not clear; however, intraruminal VFA administration may stimulate rumen epithelial morphological development in animals (Lane and Jesse, 1997). Beauchemin and Buchanan-Smith, (1989) estimated that ruminants require adequate dietary fiber intake for normal rumen function. Rumen function is associated with adequate rumination to maintain adequate salivation and optimal pH for cellulolytic microorganisms.

Examination of rumen parameters gives rapid diagnostic test for monitoring the function of the rumen as well as the nutritional health of the animals. Ruminal pH reflects the rumen acidosis condition, while, ruminal total volatile fatty acids as indicator of ruminal fermentation pattern and energy release in animal body.

Rumen ciliate protozoa play diverse and important roles in ruminal metabolism of nutrients (Williams and Coleman, 1992), they also showed that the many kinds of protozoa present in the rumen have different metabolic function and a different influence on ruminal fermentation, hence, some may be and some may not be beneficial to the ruminant host.

The reason for the beneficial effect of protozoa may be their digestive capacity, their effect on the specific growth rate of the bacteria or some general effects on the rumen environment (Kurihara *et al.*, 1968). Several factors seem to influence the concentration and composition of the protazoal fauna in the rumen; these include composition of diet, ruminal pH, ruminal temperature, turnover rate, frequency of feeding, feeding condition of the host and host species.

The microbiology of the rumen is an extremely complex subject due to the large number of organisms present with their diverse nature, and the shifting population that result from changes in the diet of the host animal. Attia *et al.*, (1980) reported that the number and types of those organisms varies according to the consumed feed. In addition marked changes may be noted within and between animals on the same or similar diets (Church, 1975).

In view of the considerable differences in development of the ruminant stomach that given concentrate or roughage diets, an experiment was made, therefore, to examine the effect of diets containing different proportions of concentrates to roughages on the rumen fermentation, the identification, density of rumen ciliate protozoa and blood parameters in early weaned lambs.

MATERIALS AND METHODS

The experiment was carried out in Ras sudr experimental research station, desert research center, located in southern Sinai governorate, during the period from April, 2009 to October 2009.

This experiment was conducted to investigate the effect of concentrate: roughage ratio on rumen development and some blood parameters in lambs early weaning.

Management and ration ingredients of experimental rations: -

Eighteen Barki male lambs, with an average live body weight 8 kg at 60days age were used in this experiment. Animals were early weaned at 8 weeks age and randomly allocated in three groups (6 lambs each) according to body weight. Different experimental groups were supported by creep feeding ration from 3 weeks until 8 weeks age , besides dam's milk. After weaning lambs depends completely on the starter pelleted ration until 24 weeks age.

Animals in the first group (T1) were offered starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10). The second group (T2) fed on starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20). The third group (T3) was fed on starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Experimental rations were isonitrogenous and isocaloric and formulated to contain (14 %DP and 67 % TDN). Rations were offered to lambs ad libtum in pelleted from 4mm screen (Table 1). Lambs were kept in semi-opend pens, while water was freely available all the day time.

Chemical analysis:-

Chemical analysis of samples feeds were carried out according to the A.O.A.C. (1990) in Animal Nutrition Laboratory of Desert Research Center. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined according to the procedures of Goering and Van Soest (1982).

Table (1). Composition of ingredient feed rations (%) used for lambs during the whole period:-

Ingredients	T1	T2	T3
Yellow corn	39	45	49
Soybean meal	12	14	15
Wheat bran	31	13	0
Hay	10	20	30
Molasses	5	5	3
Limestone	1.5	1.5	1.5
Sodium chloride	1	1	1
Mineral mixture and vitamin	0.5	0.5	0.5
Total	100	100	100
CP	14.40	14.30	14.26
%TDN	67.23	66.99	66.47

CP and TDN were obtained by calculated
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Rumen liquor analysis: -

Rumen liquor samples were obtained every 4 weeks using stomach tube at zero time (before feeding), 3, 6 and 8 hours post feeding and filtered through two layers of gauze cloth to remove feed particles. pH was immediately measured with pH meter, then 1 ml toluene and 1ml paraphen oil were added to the strained ruminal fluid and stored in deep freeze at (-20°C) until analysis. Value of pH in the rumen liquor was determined as described by the pH meter model the pHep, ammonia nitrogen concentration (NH3-N) was determined according to A.O.A.C (1990), the total volatile fatty acids (TVFA's) was determined according to Warner (1964), total nitrogen (TN) and non-protein nitrogen (NPN) were determined by the modified semi-micro-kjeldahl digestion method according to A.O.A.C (1990) while true protein nitrogen (TP) was calculated by subtracting the non-protein nitrogen content from total nitrogen content.

Ruminal ciliate protozoa count and classification: -

The number of rumen protozoa per 1 ml from rumen liquor and classification of the types of rumen protozoa were determined every 4 weeks in rumen liquor samples at zero time (before feeding), 3, 6 and 8 hours post feeding. The collected contents were immidately flittered through one layer of gauze, then fixed and stained with 4 times volume of methyl-green formalin saline solution as described by Ogimoto and Imai (1981) (100 ml formaldehyde 35 % ,900 ml distill water , methyl-green 0.6 g and sodium chloride 0.8 g), then stoked in dark place until examination.

After gentle mixing of fixed rumen liquor sample, one drop was poured on hemocytometer slide, covered with a cover slip and examined under a light microscope for identification of genera and species according to the description published by Dehority (1993).

The number of rumen protozoa per 1ml was calculated as follow:

Calculation: - number of protozoa /1 ml rumen liquor = N*5*10*4

Where:- N = count the number of protozoa in one large corner square of White Blood Cell.

Sampling of blood: -

Lambs from each treatment were used monthly to obtain 12 ml blood from the jagular vein at zero time of feeding, 4 and 8 hours post-feeding for five sequenced months. Blood samples for lambs were left to coagulate at room temperature, then centrifuged at 4000 Xg for 15 min to separate serum and kept it frozen at -20°c till analyzed for: -

The total proteins value was determined using electronic apparatus, albumin value was determined according to Doumas and Biggs (1971), globulin was obtained by substracting the albumin values from the

total proteins concentration. Serum samples were analyzed to determine urea according to Patton and Crouch (1977). Glucose and triglyceride were determined according to Trinder, (1969),

Statistical analysis: -

General linear model procedure was used for statistical analysis through SAS software (SAS, 1998), the used design was two-way analysis, and the model was: -

$$Y_{iej} = \mu + T_i + M_e + I_j + TM_{ie} + TI_{ij} + e_{iej}$$

Where: - Y_{iej} = experimental observation

μ = general mean

T_i = effect of treatment (i = 1, 2, 3)

M_e = effect of age (e = 8, 12, 16, 20, 24 weeks)

I_j = effect of time of sampling (j=0, 3, 6, 8)

TM_{ie} =effect of interaction of treatment and age

TI_{ij}=effect of interaction of treatment and time of sampling

e_{ij} = experimental error

Duncan's multiple tests were applied for comparison of means (Duncan, 1955).

RESULTS AND DISCUSSION

Chemical composition:-

The data of chemical composition of the three experimental rations are insulated in Table (2), it was important to show that T3 had the lowest value of organic matter, although it had the highest value of crude fiber and its fraction.

Table (2). Chemical analysis of experimental rations used for lambs during the whole period (% on DM basis):-

Nutrient	T1	T2	T3
Proximate analysis, DM basis:			
DM	90.59	91.17	91.02
OM	91.79	91.23	90.04
CP	14.00	13.98	13.98
CF	7.40	9.47	11.61
EE	3.44	2.82	2.10
ASH	8.22	8.78	8.78
NFE	66.94	64.68	63.53
Fiber fraction:			
NDF	37.44	34.04	39.68
ADF	11.50	13.85	15.68
ADL	2.48	3.35	4.94
Hemi-cellulose *	14.82	10.72	32.7
Cellulose**	7.33	8.45	9.89

Hemicellulose = NDF-ADF (neutral detergent fiber – acid detergent fiber)*
*Cellulose** = ADF- ADL (acid detergent fiber – acid detergent lignin)*
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Effect of treatments on rumen liquors parameters: -

Data of ruminal parameters values of the different experimental treatments are illustrated in Tables (3, 4, 5, 6&7). The data of Tables (3&4) indicated that ruminal pH values were slightly affected by treatments. Comparison among the experimental treatments during the whole period (Table 3) indicated that there was a significant difference among T1, T2 and T3 at 8 weeks, while the difference between T2 and T3 was not significant. Although the difference among the three treatments was not significant at 24 weeks. Also, the data showed that T1 had the lowest value of ruminal pH at 8 weeks (5.64), while the lowest value at 24 weeks was for T3 (6.20).

Table (3). Ruminal parameters of sheep for the whole period:-

Items	period	T1	T2	T3
pH	8 wk	5.64^{b}±0.20	5.98^{a}±0.16	5.89^{a}±0.18
	12 wk	6.05^{b}±0.13	6.33^{a}±0.18	6.20^{b}±0.13
	16 wk	5.64^{b}±0.20	5.96^{a}±0.15	5.88^{a}±0.18
	20 wk	6.05^{b}±0.13	6.16$^{a b}$±0.13	6.28^{a}±0.12
	24 wk	6.22^{a}±0.08	6.23^{a}±0.11	6.20^{a}±0.12
TVFA's	8 wk	5.02^{c}±0.18	5.21^{b}±0.201	5.33^{a}±0.22
mg %	12 wk	6.10^{c}±0.17	7.11^{b}±0.22	7.46^{a}±0.21
	16 wk	8.44^{c}±0.11	8.81^{b}±0.18	9.83^{a}±0.23
	20 wk	9.50^{c}±0.13	9.97^{b}±0.16	10.40^{a}±0.16
	24 wk	10.65^{c}±0.23	11.44^{b}±0.25	12.12^{a}±0.27
Ammonia-N	8 wk	5.95^{b}±0.18	5.97^{b}±0.17	6.02^{a}±0.18
mg %	12 wk	6.74^{b}±0.18	6.67^{b}±0.14	7.01^{a}±0.13
	16 wk	7.89^{b}±0.20	7.98^{b}±0.14	8.20^{a}±0.12
	20 wk	10.02^{c}±0.24	10.75^{b}±0.26	11.55^{a}±0.30
	24 wk	13.75^{c}±0.30	14.85^{b}±0.29	15.54^{a}±0.28
	8 wk	13.09^{a}±0.40	13.14^{a}±0.38	13.25^{a}±0.40
NPN	12 wk	14.83^{b}±0.39	14.67^{b}±0.31	15.42^{a}±0.30
mg %	16 wk	17.36^{b}±0.44	17.56^{b}±0.30	18.05^{a}±0.26
	20 wk	22.05^{c}±0.54	23.65^{b}±0.57	25.41^{a}±0.67
	24 wk	30.25^{c}±0.66	32.67^{b}±0.63	34.20^{a}±0.61
Total nitrogen	8 wk	19.77^{a}±0.60	19.84^{a}±0.58	20.01^{a}±0.60
mg %	12 wk	22.39^{b}±0.59	22.16^{b}±0.47	23.29^{a}±0.45
	16 wk	26.22^{b}±0.67	26.52^{b}±0.45	27.26^{a}±0.40
	20 wk	33.30^{c}±0.81	35.71^{b}±0.87	38.38^{a}±1.01
	24 wk	45.69^{c}±0.99	49.34^{b}±0.96	51.64^{a}±0.92
True protein	8 wk	6.67^{a}±0.20	6.70^{a}±0.20	6.76^{a}±0.20
mg %	12 wk	7.56^{b}±0.20	7.48^{b}±0.16	7.86^{a}±0.15
	16 wk	8.85^{b}±0.23	8.95^{b}±0.15	9.21^{a}±0.13
	20 wk	11.24^{c}±0.27	12.06^{b}±0.29	12.96^{a}±0.34
	24 wk	15.43^{c}±0.33	16.66^{b}±0.32	17.44^{a}±0.31

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Comparison among the experimental treatments throughout the different sampling periods (Table 4) showed slight increase with age from 8 to 24 weeks of age, the lowest value was at 8 and 16 weeks of age, while the highest value was at 24 weeks of age.

The data of Table (5) indicated that the lowest value was for lambs fed ration with 90:10 concentrate: roughage ratio (T1) (5.92) while the difference between lambs fed ration with 80:20 concentrate: roughage

ratio (T3) and lambs fed ration with 70:30 concentrate: roughage ratio (T2) was not significant being, 6.13, 6.09, respectively.

Table (4). Overall mean of ruminal parameters of sheep for the whole period:-

Items	8 weeks	12 weeks	16 weeks	20 weeks	24 weeks
PH	$5.84^{b}\pm0.10$	$6.19^{a}\pm0.09$	$5.83^{b}\pm0.10$	$6.16^{a}\pm0.07$	$6.22^{a}\pm0.06$
TVFA's mg %	$5.19^{e}\pm0.11$	$6.89^{d}\pm0.15$	$9.03^{c}\pm0.14$	$9.95^{b}\pm0.11$	$11.40^{a}\pm0.17$
Ammonia-N mg %	$5.98^{e}\pm0.10$	$6.80^{d}\pm0.09$	$8.02^{c}\pm0.09$	$10.77^{b}\pm0.19$	$14.71^{a}\pm0.20$
NPN mg %	$13.16^{e}\pm0.22$	$14.97^{d}\pm0.20$	$17.66^{c}\pm0.20$	$23.71^{b}\pm0.41$	$32.37^{a}\pm0.45$
Total nitrogen mg %	$19.87^{e}\pm0.33$	$22.61^{d}\pm0.30$	$26.67^{c}\pm0.30$	$35.80^{b}\pm0.61$	$48.89^{a}\pm0.68$
True protein mg %	$6.71^{e}\pm0.11$	$7.63^{d}\pm0.10$	$9.00^{c}\pm0.10$	$12.09^{b}\pm0.21$	$16.51^{a}\pm0.23$

Means with different litters with each row are significantly different (P<0.01).

Table (5). Overall mean of ruminal parameters of all treatments of sheep groups:-

Items	T1	T2	T3
PH	$5.92^{b}\pm0.036$	$6.13^{a}\pm0.036$	$6.09^{a}\pm0.036$
TVFA's mg %	$7.94^{c}\pm0.018$	$8.50^{b}\pm0.018$	$9.03^{a}\pm0.018$
Ammonia-N mg %	$8.87^{c}\pm0.027$	$9.24^{b}\pm0.027$	$9.66^{a}\pm0.027$
NPN mg %	$19.52^{c}\pm0.060$	$20.34^{b}\pm0.060$	$21.27^{a}\pm0.060$
Total nitrogen mg %	$29.47^{c}\pm0.091$	$30.71^{b}\pm0.091$	$32.12^{a}\pm0.091$
True protein mg %	$9.95^{c}\pm0.030$	$10.37^{b}\pm0.030$	$10.84^{a}\pm0.030$

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Data presentation (Table 6) showed that ruminal pH decreased by feeding for all treatments, it seems that the difference between T1and T3 was not significant, also the difference between T2and T3 was not significant at 0, 3 and 8 hours of feeding, while the difference among all treatments was not significant at 6 hours post feeding. The highest ruminal pH value was recorded for lambs fed ration with 80:20 concentrate: roughage ratio (T2) at zero hr time of feeding (6.86), whereas, the lowest one was that for lambs fed ration with 90:10 concentrate: roughage ratio (T1) at 3 hr post-feeding (5.59).

The data of overall means of ruminal pH at the different sampling times in Table (7) clearly showed that the ruminal pH values were higher (P<0.01) before feeding then it decreased at 3 hours post feeding then it gradually increased again. The highest value (P<0.01) was at zero time before feeding (6.74) followed by 8 hours (5.93), whereas, the lowest one (P<0.01) was recorded at 3 hr post-feeding (5.72). This can be related to ruminal fermentation process by rumen microorganisms which took place on the soluble carbohydrates very soon producing more propionate, decreasing pH value. While fermentation of the structural carbohydrates needs more time producing more acetate delaying the decreased pH value. These results are in agreement with those obtained by El-Ashry *et al.*, (1997) who reported that the minimum pH values were observed at 3 hrs post feeding (ranged between 6.29 and 6.83) and tended to increase at 6 hrs post feeding. The reduction of rumen pH after feeding could be attributed to the major role of protozoa in slowing down the fermentation by ingesting starch grains and taking up soluble sugars and converting them to storage polysaccharides.(Williams and Coleman, 1997).

Comparison among the experimental treatments (Table3) throughout the different periods showed significant (P<0.01) increase in total volatile fatty acids values with age from 8 to 24 weeks age, the lowest value was at 8 weeks age for T1 (5.02 mg/100 ml R.L) while the highest value was at 24 weeks age for T3 (12.12 mg/100 ml R.L).

Data of Table (4) indicated highly significant (P<0.01) increase in total volatile fatty acids concentration in all lambs groups with progressed age. It seems that the lowest TVA'S concentration was at 8 weeks age

(5.19 mg/100 ml R.L) then it gradually increased until reached the maximum value at 24 weeks age (11.40 mg/100 ml R.L).

Data obtained in Table (5) showed that ruminal total volatile fatty acids values (mg/100 ml R.L) were significantly (P<0.01) affected by treatments. It seems that during the whole period T3 had the highest value followed by T2 while the lowest value was for T1, the values were 9.03, 8.50 and 7.94 mg/100 ml rumen liquor (RL) for T3, T2, and T1; respectively.

Table (6). Ruminal parameters by time of sampling from sheep:-

Items	period	T1	T2	T3
pH	0	6.59^{b} ±0.07	6.86^{a} ±0.08	6.78^{ab} ±0.04
	3	5.59^{b} ±0.09	5.85^{a} ±0.05	5.74^{ab} ±0.10
	6	5.69^{a} ±0.13	5.81^{a} ±0.09	5.86^{a} ±0.09
	8	5.80^{b} ±0.13	6.01^{a} ±0.08	5.99^{ab} ±0.09
TVFA's mg %	0	7.30^{c} ±0.61	7.68^{b} ±0.64	8.13^{a} ±0.68
	3	8.62^{c} ±0.60	9.38^{b} ±0.63	9.93^{a} ±0.68
	6	8.11^{c} ±0.54	8.68^{b} ±0.55	9.31^{a} ±0.63
	8	7.74^{c} ±0.50	8.28^{b} ±0.53	8.74^{a} ±0.57
Ammonia-N mg %	0	7.86^{c} ±0.72	8.40^{b} ±0.82	8.72^{a} ±0.87
	3	9.72^{c} ±0.83	10.07^{b} ±0.94	10.49^{a} ±1.01
	6	9.13^{c} ±0.77	9.45^{b} ±0.90	9.91^{a} ±0.96
	8	8.77^{c} ±0.68	9.06^{b} ±0.82	9.54^{a} ±0.89
NPN mg %	0	17.30^{c} ±1.59	18.48^{b} ±1.80	19.20^{a} ±1.92
	3	21.39^{c} ±1.83	22.16^{b} ±2.06	23.08^{a} ±2.23
	6	20.08^{c} ±1.69	20.79^{b} ±1.99	21.80^{a} ±2.11
	8	19.29^{c} ±1.50	19.93^{b} ±1.81	20.99^{a} ±1.95
Total nitrogen mg %	0	26.13^{c} ±2.39	27.90^{b} ±2.72	28.99^{a} ±2.90
	3	32.30^{c} ±2.76	33.47^{b} ±3.11	34.85^{a} ±3.37
	6	30.3^{c} ±2.55	31.40^{b} ±3.00	32.93^{a} ±3.18
	8	29.10^{c} ±2.27	30.09^{b} ±2.74	31.70^{a} ±2.95
True protein mg %	0	8.82^{c} ±0.81	9.42^{b} ±0.92	9.79^{a} ±0.98
	3	10.91^{c} ±0.93	11.30^{b} ±1.05	11.77^{a} ±1.14
	6	10.24^{c} ±0.86	10.60^{b} ±1.01	11.12^{a} ±1.07
	8	9.84^{c} ±0.77	10.16^{b} ±0.92	10.70^{a} ±1.00

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Table (7). Overall mean of ruminal parameters by time of sampling from sheep:-

Items	0 hour	3 hours	6 hours	8 hours
PH	6.74^{a}±0.042	5.72^{c}±0.042	5.79^{c}±0.042	5.93^{b}±0.042
TVFA's mg %	7.70^{d}±0.021	9.31^{a}±0.021	8.70^{b}±0.021	8.25^{c}±0.021
Ammonia-N mg %	8.33^{d}±0.031	10.09^{a}±0.031	9.49^{b}±0.031	9.12^{c}±0.031
NPN mg %	18.32^{d}±0.069	22.21^{a}±0.069	20.89^{b}±0.069	20.07^{c}±0.069
Total nitrogen mg %	27.67^{d}±0.105	33.54^{a}±0.105	31.55^{b}±0.105	30.31^{c}±0.105
True protein mg %	9.34^{d}±0.035	11.33^{a}±0.035	10.65^{b}±0.035	10.23^{c}±0.035

Means with different litters with each row are significantly different (P<0.01).

Also, The data from Table (6) showed that ruminal TVFA's values increased at 3 hours after feeding more than before feeding then it gradually decreased by the time of sampling, the lowest ruminal TVFA's value was

recorded for T1 at zero hr time of feeding (7.30 mg %), whereas, the highest one was that for T3 at 3 hr post-feeding (9.93 mg %).

The overall means of ruminal total volatile fatty acids at the different sampling times in Table (7) clearly showed an increase ($P<0.01$) in ruminal TVFA's concentration, reached the highest ($P<0.01$) value at 3hr post-feeding (9.31 mg %) and then decreased gradually at 6 hours post feeding (8.70 mg %) and at 8 hours post feeding (8.25 mg %) to reach the lowest value ($P<0.01$) at zero time pre feeding (7.70 mg %). These results are agreed with the findings of those obtained by El-Ashry *et al.*, (1997) who reported that the maximum concentration of total VFA's were observed at 3 hrs post feeding then tended to decrease after 6 hrs. Elliott and Read (1968) showed that different roughage percents in the ration (5, 20, 25 or 50%) gave wide differences in molar proportions of VFA. Moreover, acetic acid percentage increased from 38 to 60% when roughage increased from 5 to 50%.

The present results indicated that TVFA's showed a reverse trend of pH thus the rumen pH in general decreased with increasing the TVFA's concentration. Also, Fouad, (1991) reported that concluded that the rumen pH in general decreased with increasing the TVFA's concentration in lambs rumen. Variation in rumen pH might be responsible for the changes in other ruminal metabolites. He found that the changes in the rumen pH affected microorganisms activates and consequently the mutability concentrations.

Data of the five Tables (3, 4, 5, 6 & 7) indicated that ruminal ammonia nitrogen and Non protein nitrogen take the same trend. The values in Table (3) showed significant ($P<0.01$) difference in ruminal ammonia nitrogen and non-protein nitrogen among all treatments during the whole period, also, showed a significant increase in ruminal ammonia nitrogen and non-protein nitrogen concentration (mg/100 ml R.L) with progressed age. The lowest concentration was for T1 at 8 weeks age, being 5.95 and 13.09 mg % for ruminal ammonia nitrogen and non-protein nitrogen, respectively. While, the highest concentration was for T3 at 24 weeks age, being 15.54 and 34.20 mg % for ruminal ammonia nitrogen and non-protein nitrogen, respectively.

Results obtained in Table (4) showed gradual increase of ruminal ammonia-N and NPN during the whole period, it showed gradual increase with progressed age as that the minimum value was at 8 weeks age (5.98 and 13.16 mg % for ruminal ammonia and ruminal NPN; respectively) then it increased gradually to reach the maximum value at 24 weeks age (14.71 and 32.37 mg % for ruminal ammonia and ruminal NPN; respectively).

From the data of Table (5) it seems that T3 followed by T2 significantly increased ($P<0.01$) ruminal ammonia and non protein nitrogen (mg/100 ml R.L) more than T1, the values were 9.66, 9.24 and 8.87 mg %, respectively, for ruminal ammonia, and 21.27, 20.34 and 19.52 mg/100 ml R.L; respectively, for non protein nitrogen.

Obtained data of Table (6) presented a significant difference in ruminal ammonia nitrogen and non-protein nitrogen concentration among all treatments in different sampling time. The lowest value was for T1 at 8 weeks age, being, 7.86 and 17.30 mg % for ruminal ammonia nitrogen and non-protein nitrogen, respectively. While the highest value was for T3 at 24 weeks age, being, 9.54 and 20.99 mg % for ruminal ammonia nitrogen and non-protein nitrogen, respectively.

The data of Table (7) indicated that the comparison among the experimental treatments throughout the different sampling times showed an increase of ruminal ammonia-N and NPN after feeding to reach the maximum value at 3hr post-feeding (10.09& 22.21 mg %) for ruminal ammonia and ruminal NPN; respectively) then decreased gradually to the minimum value at zero time of feeding (8.33, 18.32mg/100 ml R.L for ruminal ammonia and ruminal NPN; respectively). This increase in NH3-N concentration with post-feeding times may be related to degradation of dietary degradable protein.

The current results of ruminal ammonia may be attributed to the presence of rumen protozoa as they play an important role in the digestion of protein (Euge'ne *et al.*, 2004) and the formation of the end products of ruminal fermentation (Ushida and Jouany, 1996 and Seng, *et al.*, 2001) demonstrated a highly significant reduction of rumen ammonia nitrogen concentration when sheep were defaunated from protozoa. Moreover, Hristove, *et al.*, (2001) showed that completely eliminated protozoa reduced ammonia concentration by 60% compared with untreated control in cattle fed medium-or high- concentrate barley based diets.

From the data of the five Tables (3,4,5,6,&7) It seems that ruminal true protein nitrogen concentration was the seem trend of ruminal total nitrogen, Data presented in Table (3) showed that the lowest (P<0.01) value for total nitrogen and true protein nitrogen during the whole period was for T1 at 8 weeks age (19.77 and 6.67 mg % for total nitrogen and true protein nitrogen, respectively) while the highest (P<0.01) one was for T3 at 24 weeks age (51.64 and 17.44mg % for total nitrogen and true protein nitrogen, respectively). Data of Table (4) showed that total nitrogen and true protein nitrogen values were increased (P<0.01) by the progressed age.

The data of Table (5) also showed significant deference (P<0.01) among treatments, T3 had the highest values of total nitrogen and true protein nitrogen (32.12 and 10.84 mg %, respectively) followed by T2 (30.71 and 10.37 mg %) then T1 (29.47 and 9.95 mg %). Presented data of Table (6) clearly showed significant difference among all treatments during different sampling time, it seems that T1 had the lowest values while T3 had the highest values at all different sampling time. data of Table (7) indicated that ruminal total nitrogen and true protein nitrogen increased (P<0.01) at 3 hours post feeding then decreased gradually at 6 hours post feeding followed by 8 hours post feeding and the lowest value was at zero time pre feeding.

Effect of treatments on ruminal ciliate protozoa count and ratio: -

Tables (8, 9, 10, 11 & 12) and Curves (1, 2&3) presented the identification of ruminal protozoa species and their density in the rumen liquor [count ($x10^4$ cell/ml rumen liquor) and % ratio] during all different samples time for the whole period for all treatments. Six genera with 16 species and 6 subspecies or formae of ruminal protozoa in lambs were identified in sheep in this study, these generas (genus) are *Entodinum spp.* [*E. nanellum, E. simplex, E.exigum, E. caudatum, E.bursa, E. minimum, E. triacum* and *E. dubardi*], *Dasytrachia rummantium, Isotrchia spp.* [*I. intestinalis* and *I. prostoma*], *Diplodinum anisacanthum, Polyolastron multivesiculatum and Ophryoscolox spp.* [*O. caudatus* and *O. purkynjei*]. No evidence was indicated the presence of *Epidinum spp.* at the whole period of the experiment (from the age of 8 weeks to the age of 24 weeks) which may be excite in the rumen of the lambs after this age or may be the ruminal pH of these lambs was not suitable for the growth of *Epidinum sp* at this age or may be this absence is due to the ingredients of the rations.

As for total ruminal protozoa count ($x10^4$ cell /ml rumen liquor) and ratio of sheep groups for the whole period the data of Tables (8&9) and curve (1) showed that the difference among the five months were highly (p<0.01) significant, the values of ruminal protozoa count and ratio were increased gradually by time progressed from 8 weeks to reach the maximum value at the 24 weeks. The total count was 611.25 $x10^4$ cell/ml rumen liquor at the first month then it increased gradually to be 4338.47 $x10^4$ cell/ml rumen liquor at the fifth month, also all ruminal protozoa species were in the seem trend of increasing by time progressed like total count . It seems that *Entodinum spp.* appeared most frequently in all examined lambs, the composition rate of *Entodinum* had the highest (P<0.01) values of number and ratio for the whole period, which value was over 83% then *Diplodinum* (over 3.5%) and *Ophryoscolox spp.* (over 2.77%) were second dominants.

The present data showed no appearance for *Diplodinum* and *Polyolastron spp* in all lambs groups at age of 8 weeks, while the two species start appearance at the age of 12 weeks thus ruminal pH at this time is not adequate for its growth, although other species were start appearance from the age of 8 weeks.

Data presented in Table (10) indicated that there was significant (p<0.01) deference among the three lambs groups, lambs fed ration contained concentrate: roughage ratio 80:20 (T2) had the highest total ciliate densities of ruminal protozoa followed by lambs fed ration contained concentrate: roughage ratio 70:30 (T3) then lambs fed ration contained concentrate: roughage ratio 90:10 (T1) being 2316.41, 2199.91 and 1945.70 $x10^4$ cell/ml rumen liquor for T2, T3 and T1; respectively. These values considered as normal level in rumen (Hungate, 1966). Also, *Entodinum sp.* and *Isotrchia sp.* were the same trend with total count, while, T3 had the highest count of *Dasytrachia, Diplodinum* and *Polyolastron sps.* Although, T1 was higher in *Ophryoscolox sps.* Count, it was lower in the count of *Entodinum, Dasytrachia* and *Isotrchia sps.* then T2 and T3.

Data of Curve (2) showed significant (p<0.01) deference in overall means of protozoa ratio of all sheep groups, this data presented that *Entodinum sp.* ratio was higher in T2 followed by T1 then T3 being 87.26%, 84.25% and 83.10% ; respectively. While, T3 had the highest ratio of *Dasytrachia, Isotrchia, Diplodinum* and *Polyolastron spp.* while, the deference between T3 and T2 was not significant in *Isotrchia sp.* ratio, also, the deference between T3 and T1 was not significant in *Diplodinum sp.* ratio. Although, T1 was higher in *Ophryoscolox sps.* ratio (4.27%) followed by T3 (3.49%) then T2 (2.77%).

Table (8). Protozoa count x10^4 of lambs for the whole period:-

Items	Period	T1	T2	T3
Total count	8 wk	610.00^{a}±62.20	609.16^{a}±58.12	614.58^{a}±61.52
	12 wk	701.83^{b}±94.60	1049.58^{a}±84.62	1137.91^{a}±73.23
	16 wk	1509.58^{b}±102.42	2067.50^{a}±131.17	1477.91^{b}±147.00
	20 wk	2769.58^{b}±232.92	3444.16^{a}±398.59	3302.91^{a}±389.45
	24 wk	4137.50^{b}±248.95	4411.66^{b}±314.42	4466.25^{b}±338.98
Entodinum	8 wk	591.66^{a}±60.58	593.75^{a}±55.97	601.25^{a}±60.57
	12 wk	613.16^{b}±90.06	886.25^{a}±71.10	914.16^{a}±49.09
	16 wk	1184.58^{b}±88.18	1773.75^{a}±124.21	1111.66^{b}±128.86
	20 wk	2241.66^{c}±198.17	3039.58^{a}±372.44	2725.00^{b}±331.29
	24 wk	3300.00^{b}±188.36	3589.58^{a}±244.45	3602.08^{a}±260.65
Dasytrachia	8 wk	10.83^{a}±1.35	7.08^{a}±1.78	4.58^{b}±1.89
	12 wk	11.58^{b}±2.71	12.91^{b}±2.91	19.58^{a}±2.49
	16 wk	38.75^{b}±5.00	39.16^{b}±6.08	60.41^{a}±4.58
	20 wk	54.58^{c}±8.58	60.41^{b}±8.29	83.75^{a}±6.60
	24 wk	88.75^{c}±10.68	96.25^{b}±9.65	116.25^{a}±7.66
Isotrchia	8 wk	1.25^{a}±0.65	1.25^{a}±0.65	1.25^{a}±0.65
	12 wk	9.83^{b}±2.22	37.91^{a}±7.64	36.25^{a}±7.46
	16 wk	49.16^{b}±4.91	70.83^{a}±6.82	77.91^{a}±5.91
	20 wk	81.25^{c}±3.37	119.16^{a}±12.32	101.66^{b}±7.93
	24 wk	118.75^{b}±8.81	180.41^{a}±21.78	162.08^{c}±14.18
Diplodinum	8 wk	0.00	0.00	0.00
	12 wk	22.58^{b}±5.40	60.00^{a}±3.13	67.91^{a}±6.69
	16 wk	105.83^{a}±10.62	70.00^{b}±6.85	105.83^{a}±8.50
	20 wk	159.58^{a}±10.34	97.08^{b}±6.81	155.83^{a}±27.81
	24 wk	174.58^{b}±12.05	144.16^{c}±11.73	202.08^{a}±34.20
Polyolastron	8 wk	0.00	0.00	0.00
	12 wk	18.41^{c}±3.00	27.91^{b}±1.56	62.50^{a}±9.97
	16 wk	55.83^{a}±4.02	55.41^{a}±4.28	56.25^{a}±4.56
	20 wk	90.83^{b}±6.45	62.50^{c}±3.76	107.08^{a}±9.62
	24 wk	171.25^{a}±13.70	144.16^{b}±11.64	170.00^{a}±18.10
Ophryoscolox	8 wk	6.25^{a}±1.95	7.08^{a}±2.41	7.50^{a}±2.50
	12 wk	26.25^{a}±3.88	24.58^{a}±5.05	37.50^{a}±4.45
	16 wk	75.41^{a}±7.59	58.33^{b}±5.51	65.83^{ab}±5.60
	20 wk	141.66^{a}±22.65	65.41^{b}±3.96	129.58^{a}±18.641
	24 wk	284.16^{a}±28.87	257.08^{b}±41.91	213.75^{c}±30.23

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1. 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2. 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3. 70:30).

Table (9). overall mean of ruminal Protozoa ratio of lambs groups for the whole period:-.

Items	8 weeks	12 weeks	16 weeks	20 weeks	24 weeks
Total count	611.25^{e}±34.00	963.11^{d}±57.11	1685.00^{c}±85.21	3172.22^{b}±201.57	4338.47^{a}±171.70
Entodinum	595.55^{e}±33.13	804.5^{d}±46.46	1356.66^{c}±81.74	2668.75^{b}±182.27	3497.22^{a}±132.86
Dasytrachia	7.50^{e}±1.04	14.69^{d}±1.63	46.11^{c}±3.41	66.25^{b}±4.90	100.41^{a}±5.63
Isotrchia	1.25^{e}±0.37	28.00^{d}±4.15	65.97^{c}±3.92	100.69^{b}±5.53	153.75^{a}±9.90
Diplodinum	0.000^{e}	50.16^{d}±4.47	93.88^{c}±5.70	137.50^{b}±10.98	173.61^{a}±12.97
Polyolastron	0.000^{e}	36.27^{d}±4.68	55.83^{c}±2.41	86.80^{b}±5.02	161.80^{a}±8.52
Ophryoscolox	6.94^{e}±1.30	29.44^{d}±2.70	66.52^{c}±3.73	112.22^{b}±11.13	251.66^{a}±19.78

Means with different litters with each row are significantly different (P<0.01).

Table (10). Overall mean of protozoa count x10^4 of all lambs groups:-

Items	T1	T2	T3
Total count	1945.70^{c}±189.01	2316.41^{a}±213.00	2199.91^{b}±216.18
Entodinum	1586.21^{c}±148.45	1976.58^{a}±77.72	1790.83^{b}±174.69
Dasytrachia	40.90^{c}±4.76	43.16^{b}±5.10	56.91^{a}±5.78
Isotrchia	52.05^{c}±6.10	81.91^{a}±9.69	75.83^{b}±8.06
Diplodinum	92.51^{b}±9.95	74.25^{c}±6.84	106.33^{a}±12.63
Polyolastron	67.26^{b}±8.48	58.00^{c}±6.79	79.16^{a}±8.64
Ophryoscolox	106.75^{a}±14.95	82.50^{c}±14.33	90.83^{b}±11.87

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Table (11). Protozoa count by time of sampling from lambs groups:-

Items	period	T1	T2	T3
Total count	0	2240.00^{b}±400.08	2691.66^{a}±477.56	2712.00^{a}±502.72
	3	1281.66^{a}±239.91	1400.66^{a}±228.65	1138.00^{b}±212.88
	6	2000.66^{b}±358.29	2274.66^{a}±411.82	2209.66^{a}±400.25
	8	2260.46^{c}±459.63	2898.66^{a}±469.60	2740.00^{b}±457.59
Entodinum	0	1845.00^{c}±315.41	2341.33^{a}±421.84	2120.00^{c}±380.30
	3	1044.00^{b}±195.50	1218.33^{a}±194.72	923.33^{b}±174.12
	6	1652.33^{b}±283.93	1905.33^{a}±327.28	1827.66^{a}±339.66
	8	1803.53^{c}±351.72	2441.33^{a}±378.22	2292.33^{b}±380.18
Dasytrachia	0	30.66^{b}±6.35	31.66^{b}±7.58	68.00^{a}±13.50
	3	28.66^{b}±4.86	28.66^{b}±6.80	36.66^{a}±7.14
	6	36.33^{b}±7.04	39.33^{b}±7.91	56.00^{a}±10.34
	8	67.93^{b}±14.01	73.00^{a}±13.51	67.00^{b}±13.30
Isotrchia	0	51.33^{b}±14.16	91.66^{a}±16.66	95.00^{a}±18.93
	3	40.66^{a}±9.10	33.66^{a}±7.01	42.66^{a}±9.08
	6	53.33^{c}±10.33	95.66^{a}±20.96	83.00^{b}±16.63
	8	62.86^{c}±14.80	106.66^{a}±24.21	82.66^{b}±16.27
Diplodinum	0	113.33^{a}±25.97	89.66^{c}±13.97	183.00^{b}±38.63
	3	67.33^{a}±12.07	48.00^{b}±7.81	51.33^{b}±8.58
	6	87.66^{a}±17.04	70.66^{b}±13.29	88.33^{a}±14.04
	8	101.73^{a}±21.75	88.66^{b}±16.52	102.66^{a}±16.29
Polyolastron	0	82.66^{b}±18.62	68.00^{c}±14.28	99.33^{a}±22.75
	3	43.66^{a}±10.13	36.66^{b}±7.26	41.00^{ab}±8.86
	6	64.00^{b}±14.49	57.00^{c}±13.83	79.33^{a}±13.03
	8	78.73^{b}±22.03	70.33^{c}±16.67	97.00^{a}±18.37
Ophryoscolox	0	117.00^{b}±27.99	69.33^{c}±13.22	146.66^{a}±36.08
	3	57.33^{a}±12.18	35.33^{b}±9.86	43.00^{b}±9.60
	6	107.00^{a}±29.56	106.66^{a}±36.85	75.33^{b}±14.84
	8	145.66^{a}±40.8	118.66^{b}±39.04	98.33^{c}±18.75

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

It seems that T2 had the lowest ratio of *Dasytrachia, Polyolastron* and *Ophryoscolox spp.* more that other two groups. Results indicated that *Entodinum sp* recorded the largest count and ratio of total among all different species of ruminal protozoa at the whole period followed by *Ophryoscolox, Diplodinum, Polyolastron, Isotrchia* then *Dasytrachia spp.*

Results of Tables (11&12) and curve (3) represented the ruminal protozoa count at different sampling times and the overall means of this values, this data clearly showed that total ruminal protozoa count decreased by feeding at 3 hours post feeding more than at zero time pre feeding then increased gradually to reach the highest value at 8 hours post feeding, the values were 2547.88, 1273.44, 2161.66 and 2633.04 $x10^4$ cell /ml rumen liquor for zero time, 3 hours, 6 hours and 8 hours post feeding; respectively. Also, the data showed that T3 had the highest total protozoa count at zero time pre feeding followed by T2 then T1 being 2712.00, 2691.66 and 2240.00 $x10^4$ cell/ml rumen liquor; respectively. Although, T2 increased ($P<0.01$) total protozoa count at 3 hours post feeding and 8 hours post feeding more than other two groups while T3 was higher in total runinal protozoa count at this two times more than T1.

Table (12). Overall mean of Protozoa count by time of sampling from lambs groups:-

Items	0 hours	3 hours	6 hours	8 hours
Total count	$2547.88^{b}\pm262.77$	$1273.44^{d}\pm129.30$	$2161.66^{c}\pm221.14$	$2633.04^{a}\pm263.96$
Entodinum	$2102.11^{a}\pm213.78$	$1061.88^{c}\pm107.81$	$1795.11^{b}\pm180.00$	$2179.06^{a}\pm212.86$
Dasytrachia	$43.44^{b}\pm6.04$	$31.33^{c}\pm3.62$	$43.88^{b}\pm4.99$	$69.31^{a}\pm7.69$
Isotrchia	$79.33^{ab}\pm9.88$	$39.00^{c}\pm4.80$	$77.33^{b}\pm9.71$	$84.06^{a}\pm10.99$
Diplodinum	$128.66^{a}\pm16.91$	$55.55^{d}\pm5.60$	$82.22^{c}\pm8.48$	$97.68^{b}\pm10.40$
Polyolastron	$83.33^{a}\pm10.81$	$40.44^{c}\pm5.00$	$66.77^{b}\pm7.91$	$82.02^{a}\pm10.93$
Ophryoscolox	$111.00^{b}\pm16.21$	$45.22^{d}\pm6.14$	$96.33^{c}\pm16.28$	$120.88^{a}\pm19.61$

Means with different litters with each row are significantly different ($P<0.01$).

It seems that T2 increased ($P<0.01$) count of *Entodinum sp* at the four times of sampling followed by T3 then T1. Also, the data indicated that *Entodinum spp* was the highest count and ratio of total among all others protozoa kinds at the four times of sampling followed by *Diplodinum, Ophryoscolox, Isotrchia, Polyolastron* then *Dasytrachia spp.*.

Also, the present results indicated that diets with concentrate: roughage ratio 80:20% and 70:30% were the best in enhancing the total ruminal protozoa count ($x10^4$ cell/ml rumen liquor) more than 90:10% ratio. These results are supported by the results of Dehority and Orpin, (1988) who reported that the diets containing between 40 to 60% concentrate will support maximal protozoa numbers with a diverse fauna containing species in most of the genera, also, they added that when high or all concentrate diets are fed and ruminal pH decreases below 6.0, numbers of protozoa decreases and primarily *Entodinium* species are absent.

Moreover, those results are in line with the findings of Franzolin and Dehority (1996) reported that *Entodinium* constituted approximately 90% of the total protozoal numbers. Also, Ivan *et al.*, (2000) reported that *Entodinium* was the most detrimental of ciliate protozoa species. Hristove *et al.*, (2001) showed that *Entodinum sp.* made up 89 and 91% of the ciliate protozoal population in cattle fed medium- or high-concentrate barley –based diets. Santra *et al.*, (1998) reported that numerically the most important group of protozoa was the small *Spirotrichs* (65.6-70.1% of the total population) which account for only 4.8 to 9.4% of protozoa cell mass in the rumen of sheep and goats. Whereas *Isotricha* and large *Spirotricha* are numerically it is fewer in number. Bhatia *et al.*, (1992) indicated that total protozoa count in rumen of camels decreased 3 hrs after feeding and increased significantly 6 hrs post-feeding.

Eadie, (1962) suggested that pH was of primary importance in the establishment and maintenance of a ruminal protozoa population. The fail in ruminal pH is generally accompanied by a decrease in protozoa concentrations, an increase in the percentage of *Entodinium* species and in some instances a complete disappearance of the protozoa (Mackie *et al.*, 1978).

The present results indicated a decreasing trend of total number, differential rumen protozoal count at 3hrs post feeding for the whole period of experiment followed by a linear increase reaching the initial values at 8 hrs post feeding, the decreasing of protozoa number after 3hr post-feeding may be related to the decreasing of ruminal pH after feeding as a result of higher TVFA's concentration, also, Concentrates provide a source of rapidly fermentable carbohydrates for ruminal microorganisms, which produce and relatively accumulate VFA's to produce microbial protein thereby reducing ruminal pH. The linear increase in rumen ciliate protozoa at 6 hrs post feeding could be ascribed to migration of rumen protozoa from the rumino-reticular

fold to the rumen. It is established that the rumen protozoa sequester to the rumen medium in response to chemical stimuli originating from the diet (Kamra *et al.*, 1991).

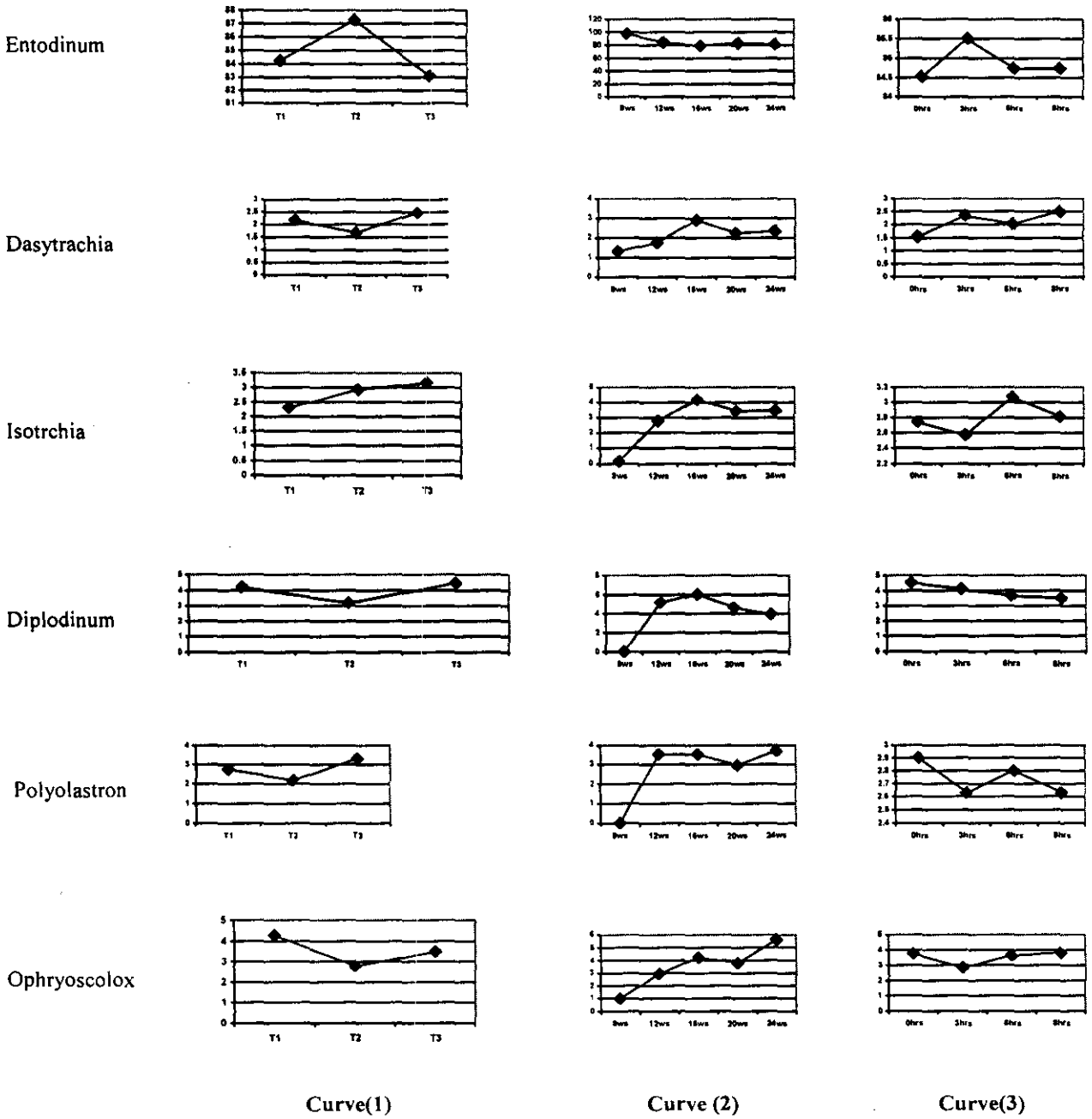

Fig. (1): Ruminal protozoa ratio of early weaning lambs as affected by age, concentrate/ roughage ratio and time of sampling:-.

Curve (1) overall mean of ruminal protozoa ratio of lamb groups as affected by concentrate/ roughage ratio.
Curve (2) overall mean of protozoa ratio of all lamb groups as affected by age.
Curve (3) overall mean of Protozoa ratio of all lamb groups as affected by time of sampling.

It is interesting to note that the numerically highest intraruminal concentrations of VFA, were occulted at 24 weeks of age of lambs, which coincided with the highest number of ruminal protozoa. These VFA and ammonia are used by ruminal protozoa to produce microbial protein to build its bodies and increasing its numbers, this process stimulates the development of rumen of these lambs. Higher total nitrogen, true protein, NPN and ammonia concentrations in the rumen of lambs fed diets with concentrate: roughage ratio 80:20% and 70:30% may be because of higher rumen microbial population, mainly rumen ciliate protozoa, contributing to rumen microbial protein synthesis.

Involving high roughage feeding for lambs developed normal rumen function characteristic. It was hypothesized that the early establishment of microorganisms would enhance rumen digestion and synthesis when high roughage rations were fed. Better fermentation of feed with in the rumen of lambs fed diets with concentrate: roughage ratio 80:20% and 70:30% could be ascribed to their higher rumen protozoa count as ciliate protozoa have a significant role in degradation of nutrients in the rumen.

Also, it is interesting to note ratio contain concentrate: roughage ratio 70:30% improved ruminal fermentation more than other rations, which indicated that the ratio of 70:30% concentrate: roughage in the rations is the best ratio for lambs from early weaning to six months. This improvement in ruminal fermentation is due to that the ratio of 70:30% concentrate: roughage had the highest density of *Entodinum sp* (which is ferment cellulose and protein), *Diplodinum sp* and *Polyolastron sp.*(which is ferment cellulose, especially that *Polyolastron sp.* can digest 50% of cellulose in the rumen) and *Dasytrachia sp.* (which is ferment volatile fatty acids) (Hungate, 1966).

Effect of treatments on blood serum parameters: -

Data in Tables (11, 12 & 13) illustrate the effect of treatments on blood serum parameters. Results in Table (11) showed that there was a significant difference ($P<0.01$) among different parameters values during the whole period. Total proteins, globulin, urea, glucose and triglyceride were gradually increased with age from the 8 to 24 weeks age. The lowest significant values were record for 8 weeks age and the highest values for 24 weeks age. While albumin values were almost the same from 8 to 24 weeks age of lambs.

Data of Table (12) indicated that the effect of the three treatments had no significant effect on total serum proteins, albumin, globulin and glucose. These results were parallel with values of CP content in the experimental rations which indicated better utilization of dietary protein through digestive tract. Kumar *et al.*, (1980) reported that serum total protein reflects the nutritional status of the animal and it has a positive correlation with dietary protein. The effect of treatments on serum urea was significant ($P<0.01$), T2 had the lowest value (37.51 g/dl) of serum urea, while T1 (40.84 g/dl) and T3 (40.12 g/dl) was almost the same values. Lambs of diet T2 and T3 had significantly lower ($P<0.01$) serum triglyceride than those of T1. The increased concentration of serum urea for animals fed low level of concentrates could be due to high urea nitrogen recycling efficiency (Badawy, 2005). Also, the data showed that glucose concentration was not affected by treatments, but, T1 and T2 had lower values than T3. Increasing serum glucose by high concentrate intake might be due to increasing the level of propionate in the rumen as reported by Provenza (1995). The present values of blood serum compounds are within the normal range for sheep and in good agreement with those obtained by El-Ashry et al., (1997). Also,These results agreed with Jakhmola and Roy (1992) who found an increase in blood glucose by increasing supplementation level (zero, 1 and 1.5 kg/head/d) being 83.5, 103.8 and 116.1 mg/dl, respectively. It is clearly that T1 had the highest values of serum triglyceride concentration at all, while T2 and T3 were not significantly differed, being 87.79, 71.29 and 74.76 mg/dl for T1, T2 and T3, respectively. These results are in agreement with those of Kouider *et al.* (1988) who found that total fat content of the plasma was dependent on the supply of nutrients.

The date of Table (13) represents the effect of time of sampling on blood compounds, total serum proteins, albumin and globulin were significantly affected by time of sampling. It is clear that the three parameters were increased at 4 and 8hrs post-feeding compared to pre-feeding values. The difference between 4hors and 8hrs was non significant being 5.98 and 5.73 g/dl; respectively for total proteins. The difference between zero time and 8hrs was non significant being 3.49 and 3.35 g/dl; respectively for albumin. Also, the difference between 4hrs and 8hrs was non significant being 2.33 and 2.38 g/dl; respectively for globulin.

Also, the data indicated that serum urea nitrogen was significantly affected by time of sampling, the values were high at zero time then it decreased ($P<0.01$) by progressed time of feeding being 48.78, 36.62 and 33.07

g/dl for zero, 4 and 8hrs; respectively. Glucose values were significantly affected by time of sampling, the values were increased at 4 hrs post-feeding compared to pre-feeding values, the highest value was at 4hrs post feeding, being 81.23, 91.52 and 85.44 g/dl for zero time, 4hrs and 8hrs; respectively. Serum triglyceride was significant (P<0.01) increased by progressed time of feeding, the highest value was at 8hrs post feeding, being 69.31, 81.84 and 82.70 g/dl for zero time before feeding, 4hrs and 8hrs; respectively.

Table (11). Blood serum compounds of lambs for the whole period:-

Items	8 weeks	12 weeks	16 weeks	24 weeks
Total protein g/dl				
Albumin g/dl				
Globulin g/dl				
Urea N mg/dl			42.05 ±1.27^{b}	
Glucose mg/dl				
Triglyceride mg/dl			0^{a}	2^{b}

Means with different litters with each row are significantly different (P<0.01).

Table (12). Blood serum compounds as affected by treatments:-

Items	T1	T2	T3
Total proteins g/dl	5.92 ± 0.13	5.78± 0.13	5.64± 0.14
Albumin g/dl	3.51 ± 0.05	3.66 ± 0.07	3.32 ± 0.08
Globulin g/dl	2.41 ± 0.14	2.12 ± 0.13	2.32 ± 0.14
Urea mg/dl	40.84 ± 1.65^{a}	37.51 ± 1.77^{b}	40.12 ± 1.47^{a}
Glucose mg/dl	86.48 ± 1.73	86.27 ± 1.65	85.45 ± 2.33
Triglyceride mg/dl	87.79 ± 4.06^{a}	71.29± 3.55^{b}	74.76 ±3.83^{b}

Means with different litters with each row are significantly different (P<0.01).
(T1) starter pelleted consists of 90 % concentrate and 10% roughage (T1, 90:10).
(T2) starter pelleted consists of 80 % concentrate and 20% roughage (T2, 80:20).
(T3) starter pelleted consists of 70 % concentrate and 30% roughage (T3, 70:30).

Table (13). Blood serum compounds as affected by time of sampling:-

Items	Zero hours	4 hours	8hours
Total proteins g/dl	5.62±0.13^{b}	5.98±0.13^{a}	5.73±0.14^{ab}
Albumin g/dl	3.49±0.06^{b}	3.65±0.07^{a}	3.35±0.08^{b}
Globulin g/dl	2.13±0.15^{b}	2.33±0.14^{ab}	2.38±0.13^{a}
Urea mg/dl	48.78±1.34^{a}	36.62±1.41^{b}	33.07±1.54^{c}
Glucose mg/dl	81.23±1.76^{b}	91.52±1.96^{a}	85.44±1.88^{b}
Triglyceride mg/dl	69.31±3.29^{b}	81.84±4.09^{a}	82.70±4.08^{a}

Means with different litters with each row are significantly different (P<0.01).

CONCLUSION

Involving high roughage feeding for lambs, especially the ratio contains concentrate: roughage ratio 70:30% enhances the fermentation of rumen parameters such that it had the lower ruminal pH among all treatments at 24 weeks age, it increased ruminal total volatile fatty acids, ammonia nitrogen, non-protein nitrogen, total nitrogen and true protein nitrogen concentration. Also, this ration had the highest deferential of rumen ciliate protozoa. Although that; this ratio had the highest value of crude fiber and its fraction and not significantly differed in blood parameters from other rations. This development in rumen fermentation may be expected to be reveres on the performance of lambs fed on this ration including live body weight and average

daily gain. Also, this ratio (70:30%) may be reduced the cost of feeding more than other ratios. So we recommended using high roughage feeding for early weaned lambs form the age of 8 to 24 weeks.

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تخمرات الكرش، بروتوزوا الكرش الهدبية و بعض قياسات الدم فى الحملان المفطومة مبكراً المغذاه على نسب مختلفة من المركز و الخشن.

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تم دراسة تخمرات ، تعريف وكثافة برتوزوا الكرش و بعض قياسات الدم فى 18 ذكر من حملان البرقى المفطومة مبكراً بمتوسط وزن جسم حى 8 كجم على عمر 60 يوم. تم توزيع الحملان عشوئياً فى ثلاث مجاميع (6 حمل فى كل مجموعة) حسب وزن الجسم. تم تغذية الحملان فى الثلاث مجاميع على مكعبات بادئ تتكون من نسب مختلفة من المركز و المالئ (معاملة (1) بنسبة 90 : 10 ، معاملة (2) بنسبة 80 : 20 و معاملة (3) بنسبة 70 : 30 %). و قد استمرت التجربة لمدة 16 أسبوع.

وقد أظهرت النتائج الرئيسية أن قيم الرقم الهيدروجينى للكرش قد تأثرت قليل بالمعاملات. بينما تأثرت تركيزات كل من الاحماض الدهنية الطيارة الكلية بالكرش و نتروجين أمونيا الكرش و نتروجين الكرش الكلى و النتروجين غير البروتينى بالكرش و نتروجين البروتين الحقيقى بالكرش تأثراً معنوياً بالمعاملات . ويبدو أن المعاملة الثالثة قد حصلت على أعلى القيم خلال الفترة الكلية يليها قيم المعاملة الثانية ثم قيم المعاملة الاولى. و قد كان من المهم توضيح وجود زيادة معنوية لهذه القياسات مع التقدم فى العمر من الأسبوع الثامن و حتى الأسبوع الرابع و العشرون. أيضاً فأن هذه القياسات زادت بعد 3 ساعات من التغذية بدرجة أكبر من قبل التغذية و التى تناقصت تدريجياً مع وقت العينات.

و قد تم تعريف 6 أجناس تضم16 نوع و 6 تحت نوع من بروتوزوا الكرش فى الحملان و هى:

[*Entodinum, Dasytrachia, Isotrchia, Diplodinum, Polyolastron and Ophryoscolox sps.*].

و قد زادت قيم أعداد بروتوزوا الكرش و نسبتها تدريجياً مع تقدم العمر من الأسبوع الثامن لتصل إلى أعلى قيمة عند الأسبوع الرابع و العشرون.

و لم يكن هناك تأثيراً معنوياً للمعاملات الثلاثة على بروتين السيرم الكلى و الألبيومين و الجلوبيولين و الجلوكوز. وقد زادت القيم عند 4 و8 ساعات بعد التغذية مقارنة بعينات قبل التغذية. و قد كان للمعاملة الثانية أقل قيمة ليوريا السيرم بينما كان للمعاملة الأولى و الثالثة نفس القيم تقريباً و كانت القيم عالية قبل التغذية ثم قلت بدرجة كبيرة مع تقدم الوقت من التغذية. و كان لحملان المجموعة الثانية و الثالثة قيم منخفضة معنوياً من ترايجلسريد السيرم عن قيم المجموعة الأولى ، و قد زادت القيم معنوياً مع تقدم الوقت من التغذية حيث كانت أعلى قيمة بعد 8 ساعات من التغذية.