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Some Rumen and Blood Metabolites and Total Bacterial Count in Camels

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Abstract:
Camels production is very important in the Sudan and nutrition is a main constraint for camels production due to rangeland deterioration and seasonal variations in feeds quantity and quality affecting animals health and performance. In addition modern nutritional concepts are not applied due to lack of information. Consequently, a series of experiments were conducted to furnish the required information. Three rumen fistulated Arabian camels were used to study some rumen and blood metabolites and total bacterial count (TBC). The animals were fed *Medicago sativa* (Barseem) *ad libitum* and concentrate ration at 0 (A), 3 (B) and 1.5 (C) kg daily. Rumen and blood samples were collected at different times (before feeding, during feeding and at 2, 6 and 9hrs after feeding) to study some rumen and blood metabolites and total bacterial count. Results showed that rumen pH (5.13 - 6.17) varied significantly ($P<0.05$) with time in diets A and C before and at feeding with diet B at 9hrs after feeding . Rumen ammonia - N (144.67 - 192.27) mg/l and urea (24.30 - 43.10) mg/dl, blood urea (53.0 - 65.73) mg/dl and total bacterial count (3.82 - 7.38) $\log_{10}$ CFU/ml were not significantly ($P>0.05$) affected with time and treatments and interaction between treatments and time was not significant ($P>0.05$). It is concluded that rumen pH, ammonia - N and urea, blood urea and total bacterial count were affected with feeding time at all concentrate ration levels and times for the highest and least values varied among diets as we recommend that Further studies are required on camels rumen metabolites and microbes and blood metabolites on different feeds.

Keywords: *camels, rumen, blood, metabolites, bacterial count.*

Introduction

Dairy products and meat demand and prices increased substantially in the Sudan in the last decades due to the increased human population and urbanization and improved education, income, living standards and nutritional awareness. It is vital to produce cheap and high quality meat and dairy products by improving conventional animal species performance and exploiting neglected species. Camels are attractive for meat and milk production due to high population ranking 2nd to Somalia in world population (FAO,2009). They are efficient producers of high quality milk and meat in arid and semi-arid environments where it is difficult to

rear other meat and milk producing animals (Farah *et al*., 1992). They have many valuable and distinguished products including milk, meat, hides and waber (Albert, 2002). In addition they are used for riding, racing and packing. Nutrition is a main constraint for camels production in the Sudan due to many factors. Rangeland generally deteriorated due to haphazard agricultural expansion and reduced area, successive droughts, over grazing and seasonal variations in feeds quantity and quality leading to serious shortages in the dry season affecting animals performance and health (Ali,2003). Information on camels nutrients requirements are scarce and information on cattle is used to calculate nutrients requirements. In addition modern concepts of ruminant nutrition as the new protein systems (ARC,1982) are not applied in camels nutrition . The new protein systems are based on microbial yield and feeds rumen degradation. Furthermore, information on rumen environment and microbes are scarce. Consequently, this study was conducted to furnish these information to improve camels nutrition and production.

Materials and Methods

Study Site

This study was conducted at the Central Veterinary Research Laboratory (CVRL), Animal Resources Research Corporation (ARRC), Ministry of Animal Resources, Fishers and Rangelands in Soba, Khartoum State, Sudan.

Animals

Three Arabian fistulated camels, two females and one male at 5-8 years old and 291- 383kg live body weight were used in this study. They were injected with Ivomec **(**Ivermctine) against internal and external parasites.

Housing

The animals were allocated at random to three individual pens shaded with corrugated iron sheets. The pens were 3.95 x 2.95 m in dimensions and were 3m high. Each pen has feeder and water trough.

Surgical preparation

They were fasted of feed and water for 24 hrs before the operation. The animals were fitted with the rumen fistulae in November 2015 as described by Brown *et al* . (1968).

Anaesthesia

Xylazine (2%) at 0.25 ml/100kg body weight was injected intramuscularly to sedate the animals. The animals were then anesthetized with Lidocaine (2%) for local infiltration and paravertebral nerve block.

Fistulation technique

Cannulae

The cannulae were 10.5 cm long tubes and were 4.5 cm in diameter. They were made from Teflon. They had a flang at one end to prevent it from coming outside the rumen. The other end of the cannulae was screwed to secure a cover. Ascrewed ring was used to fix each cannulae in the animals after fistulation. In addition two hard plastic rings were used to secure the cannulae in position and one was intact with the skin.

Post-operative care

To avoid the post-operative infection, the animals were injected with a broad spectrum antibiotic (Penivet Forte) intramuscularly for 7 days. The wounds were cleaned daily with Potassium Permanganate and Iodine. Pencillin powder was applied on wounds. The fistulated animals healed without problems and were ready for the experiment after 4 - 6 weeks from the surgery. The cannulae were cleaned regularly with a disinfectant.

Experiment

The three rumen fistulated camels were used in a 3×3 Latin square design. The animals were subjected to three different levels of the concentrate ration (A , B and C). The concentrate ration ingredients and

calculated composition and energy are shown in Table1**.** In addition the animals were offered minerals and vitamins blocks. Clean water was available all the time.

Treatments

A : The animals were offered air dried *Medicago sativa* (Barseem) *ad libitum* without concentrate ration (control) .

B : The animals were offered *Medicago sativa ad libitum* and 3kg concentrate ration.

C : The animals were offered *Medicago sativa ad libitum* and 1.5 kg concentrate ration.

The daily diets were fed in one meal at 7.30 am . The refusals were collected daily before the morning meal and recorded then the feed intake of each animal was determined. The experiment lasted 33 days including a 10 days adaptation period and one day for samples collection for each period. The adaptation period consisted of a 3 days changeover period followed by a 7 days adaptation period. Rumen liquor and blood samples were collected before feeding, at feeding and then after 2hrs , 6hrs and 9hrs. The rumen liquor was strained through two layers of sterilized gauze and immediately used for the determination of pH , NH_3 , urea and bacterial cell count. Blood samples were used to determine urea.

Laboratory analysis

The rumen pH was measured using a standard laboratory pH meter . Rumen NH_3 was determined as described by Tandon (1993). Rumen liquor and blood urea were analysed according to Tietz (2005) .

Bacterial count

The rumen liquor samples were strained by sterilized gauze and used for determination of bacteria cell count according to the spread plate method (Harrigan,1998) . A range of five - fold dilutions was used and an inoculum of 0.2 ml was pipetted into the surface of separate Agar plates.

Preparation of plate count Agar medium (Hi media M091 A)

Seventeen and half gm of plate count Agar were suspended in 1000 ml distilled water, which was heated to boiling to dissolve the medium completely. It was sterilized by autoclaving at 15 Lbs pressure at a temperature of 121ºc for 15 minutes, cooled to 45- 50 ºc and then mixed well and poured into sterilized petri plates . This medium consists of casein enzymic hydrolysate 5gm/L, Yeast extract 2.5 gm/L , Dextrose 1gm/L , Agar 9 gm /L and final pH at 25 ºc was adjusted 7.0 ± 0.2 . Two plates were inoculated per selected dilution . The inoculum was spread rapidly over the entire Agar surface using a flame sterilized nichrome wire bent in an L - shape . The plates were incubated in an Incubator for 24 hrs at 37°c. After incubation the plates were inoculated with the diluted samples were used for the determination of the colony count . An average of any two inoculated plates was used for the determination of the colony count of each sample . Calculation of number of the colony forming unit (CFU) :

Number of CFU/ml $= N \times 10^{n} \times 5$

N = No . of colonies on plate at the selected dilution n .

The colony counting method was used and plate counts 30 - 300 colonies were taken as colony forming unit (CFU).

Statistical analysis

The data was subjected to statistical analysis using two way analysis of variance (ANOVA) by the statistical computer programe (Stat Soft, 2011).

Table 1. The ingredients and calculated composition of the concentrate ration fed to the fistulated camels.

Ingredients	(%)
Sorghum grains (Feterita)	47
Groundnut cake	10
Wheat bran	40
Minerals / Mixed	02
Salt	01
CP	18.9
ME (MJ/Kg DM)	12.6

Results and Discussion

Table 2 shows some rumen and blood metabolites and total bacterial count in camels.

Rumen parameters

pH

The rumen pH varied significantly ($P<0.05$) with time in diets A and C before diets and feeding time on rumen pH. Similar results were reported by Orskov (1982). The increased pH in diet A from 6.07 before feeding to 6.10 at feeding showed that feeding activated rumen fermentaion due to substrare availability. The gradual decrease in pH up to 9hrs after feeding reflected that fermentation was suppressed with nutrient depletition and fermentaion end products as VFA . The highest pH during feeding was because the microbes had access to the required nutrients for optimum fermentation. Diet B rumen pH generally was lower than the other two feeds due to high concentrate ration level and more propionic acid produced. Diet B rumen pH decreased from 5.93 before feeding to 5.80 at feeding. It then decreased gradually up to 9hrs after feeding due to depleted nutrient and fermentation end products accumulation. It was highest before feeding due to high concentrate ration level and more propionic acid produced later on. The reduced pH in diet C from 6.17 before feeding to 6.0 at feeding was and at feeding with diet B at 9hrs after feeding. The treatments and interaction between treatments and time were not significant ($P>0.05$). The significant variations in rumen pH with time in diets A and C before and at feeding with diet B at 9hrs after feeding reflected effects of mainly due to feed activated rumen fermentation. The gradual decrease in pH up to 6hrs after feeding was mainly due to substrates depletition and end products accumulation. The similar pH at 6 and 9hrs after feeding showed that the changes were no critical between these two times. The highest pH before feeding as because substrates are deficient and fermentation and end products were least. The variations in highest and least pH among diets reflected variations in active fermentation and maximum end products. Diet A generally highest rumen pH at all times, reflected feed effects on fermentation with more acetic acid. Diet C highest pH before feeding and at 9hrs after feeding was due to least available nutrients for the microbes and less active fermentation. Rumen pH was never highest at any time in diet B was mainly due to the highest concentrate ration level with more propionic acid. Diet B generally least rumen pH at all times was mainly due to high propionic acid. The pH was never

the least at any time in diet A as it was fed sharp change in rumen pH with time in diet B than the other two diets was mainly due to high concentrate ration level and that reported by many workers (Ghosal *et al*., 1981 ; Baraka *et al.*,2000 ; Kamal, 2010) . The differences may be due to variations in diets, rumen fermentation and fermentation end productes.

Ammonia - N

The rumen ammonia - N varied with time in all diets, but not significantly (P>0.05). The treatments and interaction between treatments and time were not significant (P>0.05). The variatioins in rumen ammonia - N with time in all diets was mainly due to variations in substrates supply, fermentation and fermentation end products. Diet A gradual increase in rumen ammonia - N from feeding to 6hrs after feeding followed by a decrease at 9hrs after feeding reflected variations in fermentation activity and ammonia - N produced..The highest ammonia - N 6hrs after feeding reflected active fermentation peak. Diet B rumen ammonia - N fluctuations with time were associated with variations in fermentation and end products. The highest and least rumen ammonia - N times varied among diets due to substrates availability and active fermentation and end products . Diets B and C had the highest rumen ammonia - N at feeding due to concentrate ration enhancing active rumen fermentation and end products and mastication increased saliva outflow into the rumen. Diet C highest ammonia - N at generally all times was mainly due to concentrate ration. The highest rumen ammonia - N in diet C before feeding and least in diet A was mainly due to concentrate ration. The sharp change in rumen ammonia - N with time in diet B than the other two diets was due to the highest concentrate ration level. The rumen ammonia - N was in range (144.67 -192.27) mg/l. Similar findings were found by Satter and Slyter (1974). a roughage with more acetic acid. The propionic acid production. In this study the rumen pH (5.13 - 6.17) was lower than

However, it was higher than that obtained by Kamal (2010) and Baraka *et al.*(2000). Ammonia - N variations could be due to variations in feeds, rumen fermentation and fermentation end products..

Urea

The rumen urea varied with time in all diets, but not significantly (P>0.05). The treatments and interaction between treatments and time were not significant (P>0.05). The variations and fluctuations in rumen urea with time in all diets were associated with variations in rumen ammonia - N and pH and were manily associated with substrates availability and active fermentation. The variations in the highest and least rumen urea times among diets were also found for ammonia - N and pH and were mainly associated with substrates availability, active fermentation and end products accumulation. In diet A it was highest at all times compared to the other two diets. Diet B had the least rumen urea only at 9hrs after feeding. The sharp change in rumen urea with time in diet A than the other two diets was mainly due to the high roughage diet and acetic acid production. Rumen urea was (24.30 - 43.10) mg/dl differed from that reported by Kamal (2010) and Baraka *et al.*(2000). Rumen urea variations may be attributed to the variations in rumen ammonia - N, pH and fermentation. **Total bacterial count** The total bacterial count (TBC) varied with time in all diets, but not significantly (P>0.05). The treatments and interaction between treatments and time were not significant (P>0.05). The variations with time in total bacterial count in all diets was associated with the variations in all the previous parameters in rumen and blood. This was mainly due to feeds and time. The fluctuations in

(TBC) with diets were also found for rumen pH, ammonia - N , urea and blood urea. The sharp change in (TBC) with time in diet A than the other two diets was mainly due to high roughage and acetic acid produced. In this study total bacterial count was not in line with that found by Rabee *et al*. (2016) and Chaucheyras-Durand and Ossa (2014). These variations could be due to the differences in rumen ammonia - N, urea and pH and blood urea. There is no available information in camels. **Blood metabolites Blood urea** The blood urea varied with time in all diets, but not significantly (P>0.05). The treatments and interaction between treatments and time were not significant (P>0.05). The variations in blood urea with time in all diets were associated with variations in rumen ammonia - N , urea and pH which were associated with time after feeding and active rumen fermentation. The fluctuations in blood urea with time in all diets were associated with those in rumen ammonia - N , urea and pH. The highest blood urea in diet B only before feeding was due to high concentrate ration and propionic acid produced. The generally highest blood urea in diet C in all times was mainly due to moderate concentrate ration level. Diet A least blood urea before and at feeding was mainly due to high roughage. Diet B least blood urea from 2-9hrs after feeding was associated with higher concentrate ration and propionic acid. The highest blood urea in diet C 2hrs after feeding and the least in diet B at 9hrs after feeding reflected that diet C allowed optimum rumen fermentation due to moderate concentrate ration. The sharp change in blood urea with time in diet C than the other two diets. The results showed variations with many other authors (Kumar *et al*.,1961 ; Soliman and Shaker, 1967 ; Koudier and Kolb, 1982a ; Chavanne and Bone,1950). The discrepancy may be due to variations in rumen ammonia - N , urea and pH.

Conclusions

This study concludes that Diets affected on rumen microbes, pH, ammonia - N , urea and blood urea. Further studies are required on camels rumen metabolites and microbes and blood metabolites on different feeds. In addition to use available information to adopt modern ruminants nutrition systems in camels nutrition for efficient feeding and production.

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Table 2. Some rumen and blood metabolites and total bacterial count in camels.

Time	Before feeding			At Feeding			After 2 hrs			After 6 hrs			After 9 hrs			SEM	P		
Treatment	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	-	Time	Treatment	Time * Treatment
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	-	-	-	-
pH	a 6.07	ab 5.93	a 6.17	a 6.10	ab 5.80	a 6.00	ab 5.87	ab 5.67	ab 5.63	ab 5.53	ab 5.30	ab 5.40	ab 5.37	b 5.13	ab 5.40	0.25	0.003	0.383	0.999
NH_3-N mg/l	144.67	150.27	192.27	169.87	182.93	182.93	178.27	164.27	171.73	181.07	172.67	187.60	174.53	163.33	180.13	14.07	0.557	0.169	0.705
Rumen urea mg/dl	38.10	32.83	29.77	32.53	30.77	28.47	43.10	30.83	24.30	39.20	31.43	26.70	38.03	25.97	29.47	7.10	0.986	0.069	0.985
Blood urea mg/dl	58.60	60.17	58.67	57.30	58.83	65.00	56.67	53.43	65.73	55.70	53.20	58.00	60.87	53.00	61.63	6.39	0.923	0.324	0.980
TBC log_{10} CFU/ml	7.38	5.27	5.67	6.87	5.68	7.27	7.27	7.38	6.82	4.80	6.61	5.60	5.84	5.76	3.82	1.34	0.407	0.784	0.835

SEM = Standard error of means

P = Probability

بعض المستقلبات الايضية في الكرش والدم وعدد الباكتيريا الكلي في الابل.

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المستخلص

يعتبر انتاج الابل هام جدا في السودان كما ان التغذية معوق رئيسي لانتاج الابل لتدهور المراعي والاختلافات المؤسمية في كمية ونوعية الاغذية مما يؤثر علي صحة واداء الحيوانات . اضافة لعدم تبني الافكار الحديثة للتغذية لعدم توفر المعلومات . لذلك اجريت سلسلة من التجارب لتاسيس المعلومات المطلوبة . ثلاثة من الابل العربية ثبتت بها نواسير بالكرش لدراسة بعض المستقلبات الايضية في الكرش والدم والعدد الكلي للباكتيريا . تم تغذية الحيوانات علي البرسيم حد الشبع وعليقة مركزة عند 0 (A) , 3 (B) و 1.5 (C) كيلوجرام يوميا . تم جمع عينات الكرش والدم في اوقات مختلفة (قبل الاكل , اثناء الاكل وعند 2 , 6 , 9 ساعات بعد الاكل) لدراسة بعض المستقلبات الايضية وعدد الباكتيريا الكلي . اظهرت النتائج تباين الاس الهيدروجيني معنويا ($P<0.05$) مع الزمن في العليقتين أ وج قبل وعند الاكل مع العليقة ب عند 9 ساعات من تقديم العليقة . لم يكن اثر الزمن معنويا ($P>0.05$) علي كل من الامونيا (192.27 – 144.67) mg/l واليوريا mg/dl (43.10 – 24.30) في الكرش و يوريا الدم mg/dl (65.73 – 53.0) و عدد الباكتريا الكلي log_{10} CFU/ml (7.38 – 3.82) . لم يكن اثر المعاملات والتداخلات بينها والزمن معنويا ($P>0.05$) لكل عوامل الكرش ويوريا الدم . خلصت الدراسة علي ان عوامل الكرش ويوريا الدم كانت متاثرة بوقت التغذية في كل مستويات العليقة المركزة وتباينت اوقات القيم الاعلي والاقل بين المعاملات كما نوصي بمذيد من الدراسات المطلوبة علي المستقلبات الايضية في الكرش و المايكروبات و المستقلبات الايضية للدم في الابل علي الاغذية المختلفة .