ANNUAL PROGRESS REPORT OF THE ICAR ADHOC PROJECT INVESTIGATION ON THE URINARY CALCULOGENIC AND CALCULOLYTIC AGENTS IN RUMINANT AND MONOGASTRIC ANIMALS FROM 15.2.98 TO 14.2.99





DEPARTMENT OF ANIMAL NUTRITION COLLEGE OF VETERINARY AND ANIMAL SCIENCES KERALA AGRICULTURAL UNIVERSITY MANNUTHY, THRISSUR

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1.	Project.	Title	:	INVESTIGATION ON URINARY CALCULOGENIC AND CALCULOLYTIC AGENTS IN RUMINANT AND MONOGASTRIC ANIMALS
2.	Sanction	L No.	:	F. No. 8 - 8/94 ASR II dt. 20.11.96 of Asst. Director General (AN & P), I.C.A.R., NEW DELHI
3.	Report	period and No.	:	15.2.1998 to $14.2.1999$ APR $2/98 = 99$
4 🛶	Date of	start	:	15-2-1997
5.	Date of	Termination.	:	14.2.2000
6.	a. Name	of Institute/Station	:	College of Veterinary & Animal Sciences, Kerala Agricultural University MANNUTHY
	b. Divi	sion/Dept./Section	:	Department of Animal Nutrition College of Veterinary & Animal Sciences, MANNUTHY
	c. Loca	tion of work	:	Department of Arimal Nutrition and Small Animals Breeding Station
	-	rks of scientific panel previous years report	:	· · ·
7.		nical programme approved the scheme	:	
				involues the experimental

ANNUAL PROGRESS REPORT OF RESEARCH SCHEME

The technical programme involves the experimental production of urolithiasis in different groups of animals and its ameleoration by administering chemical or plant agents. The mat erials proposed to investigate the anticalculogeric/calculolytic potential are,

Chemical agents

i. Elemental Sulphur

ii. Calcium. Oxide.

iii. DL - Methionine

- iv. Vitamin D
 - v. Ammonium chloride
 - vi. Citric acid

<u>Plant agents</u>

- i. Garlic/Onion
- ii. Tamarind
- iii. Horse gram.
- iv. Banana stem

These materials will be subjected to investigation individually or in combination in suitable cases. The experimental animals will be

Ruminant	-	Goat
Monogastric	_	Rat.

The experiment will be carried out in three phases Phase I

Experimental production of urinary calculi in different groups of animals and ascertain the type of calculi formed

<u>Phase II</u>

Protecting the animals from the incidence of urinary calculi by supplementing/administering the proposed anticalculogenic agents individually or in combination along with the calculogenic ration

Phase III

Ameleoration of urinary calculi by using these anticalculo – genic agents on the affected animals and its effect on the resolution of urinary calculi which is already formed

FIRST YEAR

Phase I

A minimum of 18 animals will be required in each species for conducting the experiment. The animals will be devided into 2 groups of 9 animals each and will be maintained on a normal ration in one group and a calculogenic ration in another group for a period of over three months. Records of daily feed consumption and weekly weight gain will be maintained. Haematological data including enzyme levels and rumen volatile fatty acid levels (ruminants) will be recorded and metabolism trial will be carried out periodically to assess the metabolic variation due to calculogenic ration in the animals under the trial. At the termination of the trial the animals will be slaughtered for post mortem and histopathological studies to ascertain the formation of calculi

The gross calculi formed will be collected and will be subjected to further chemical analysis to ascertain the type of calculi experimentally produced.

SECOND YEAR

<u>Phase II</u>: A minimum of 60 animals in each species will be required for conducting the trial. Ten groups of 6 animals will be maintained on a calculogenic ration supplemented with the proposed anticalculogenic agents and will be maintained for over a period of three months. Daily records of feed consumption, body weight gain, haematological values, rumen volatile fatty acid levels (ruminants) and data on metabolism trial will be gathered to assess the metabolic role of each anticalculogenic agent and to derive conclusions therefrom supported by histopathological studies.

Screening of potential anticalculogenic agents will be done in this phase. These agents will be further subjected to investigation individually or in combination in different groups of animals for their calculolytic activity.

THIRD YEAR

Phase III

<u>Stage I</u>: A minimum of 84 animals in each species will be required for assessing the efficacy of each agents on dissol ution of urinary calculi in situ. For this, the experimental will be maintained on a calculogenic ration for a period of three months and 12 animals from each species will be slau ghtered for post mortem and histopathological studies to ascertain whether the animals maintained on the calculogenic ration developed urinary calculi

<u>Stage II</u>: The rest of the animals maintained on the calculogenic ration will be devided into different groups depending upon the potential calculolytic agents to be tested. The cal culolytic agents in the form of a supplement or extract may be administered to the animals in which calculi is already formed. Periodical slaughter and histopathological studies apart from routine haematological and metabolism data will help to assess the efficacy of calculolytic agents.

Main observation to be recorded

- 1. Feed consumption
- 2. Body weight gain.
- 3. Haematological data
- 4. Rumen: VFA values
- 5. Metabolism trial data
- 6. Histopathological observation
- b. Bechnical programme approved for the year : As above
- c. Technical programme approved for next year : As above

8. Technical personnel employed (List vacancy if any)

Name with Designation	Date of joining	Date of leaving
1. Miss. Mini, V. Research Associate	15.2.1997	• •
2. Mr. Sukkoor, Y Jr. Research Fellow	12.5.1997	••
Recruitment position	All the san filled up	ctioned posts are

		· · ·		
9.	Total out	:lay :	Rs.	10,01,225.00 for 3 years
10.	Total amo previous	year		1,75,076.00 enditure on NRC not included
11.	Total amo	ount spent/sanctioned duri	ng tì	ne year under report:
	a,	Sanctioned		
		Fay & Allowances	Rs.	77,242.00
		Contingency Recurring	Rs.	1,61,500.00
	1	Non recurring		-
		Total	Rs.	2,38,742.00
	b.	Spent		
		Pay & Allowances	Rs.	63,034.00
		Contingency recurring	Rs.	1,61,483.00
		Non recurring	Rs.	2,75,709.00
		Total	Rs.	5,00,226.00
12.	Total nur the year	mber of man months during:		24
13.	0 bjec ti v	98:		
	i.	Experimental production of	of ur:	inary calculi in animals
	ii.			the incidence of calculi. g chemical/plant agents along
	iii.			li by using these agents on resolution of calculi which
14.	Approved	technical programme		

a. For the year under report - As in item 7

b. For the next year - As in item 7

15. Progress of Research:

The first year of the project was started with the appoint ment of staff. They were being trained for the analysis of feed and biological materials. All the equipments have been procured during the early part of the second year. Experimental animals have been purchased from University Sheep & Goat Farm, Mannuthy as and when it is required

FEEDING TRIALS WITH RUMINANT

A. CALCULOGENESIS

As envisaged in the technical programme, 18 goats were selected and devided into two groups of nine animals each as uniformly as possible with regard to sex, age and weight and main tained on a normal ration in one group and a potential calculogenic ration in group two and maintained individually for a minimum period of three months. Weighed quantity of feed was given to each animal daily, their requirement being worked out on the basis of their body weight. Water was provided <u>ad. libitum</u>. Daily feed consumption record was maintained as well as weekly body weight. Biweekly hae matological data and monthly metabolism trial data on nitrogen, cal cium, phosphorus and magnesium, urine pH variation if any and extent of crystalluria in animals maintained under each group were gathered.

Calculogenic Ration

A calculogenic ration was computed by looking into the nutrient requirement, specially taken into consideration of the mineral content of the ration. In the diet, the mineral content of the control and calculogenic ration is as shown below:

	<u>Control</u>	<u>Calculogenic</u>
Calcium	0.79 %	1.22 %
Phosphorus	0.43 %	0.63 %
Magnesium	0.31 %	1'.20 %

Control ration is fortified with calcium to the extent of 1.22 %, phosphorus to the extent of 0.63 % and magnesium to the extent of 1.20 % by supplemental chemicals like calcium oxide, phos phorus pentoxide and magnesium oxide respectively

As the roughage part, guinea grass was provided and a concentrate - roughage ratio of 4:1 was always maintained through out the experimental period.

Data Collection

Data pertaining to weekly weight gain and daily feed intake were maintained throughout the course of the study and also records on R.B.C., W.B.C., Haemoglobin and Plasma protein concentrations, calcium, phosphorus and magnesium levels in blood are being estimated at fortnightly intervals in all series of experiment. Blood samples for analysis were withdrawn from the jugular vein into sterilised citrated tubes. Mineral and nitrogen balances were determined for a period of five consecutive days at the beginning of the feeding trial and at 28 days interval in all series of experiment. Thymol and sul phuric acid was used as urine preservatives.

Discussion

<u>Body weight gain</u>: Average weekly body weight gain recorded for the goats maintained under the two groups (A and B) up to 112 days of experiment is shown in Tables I and II and an increase in body weight during the period was noticed for the groups A and B, gain accrued being 8.6 and 8.4 Kg respectively indicating no appreciable difference between the groups with regard to body weight gain

Red Cell, W.B.C., Haemoglobin and Plasma Protein Concentrations : Average biweekly R.B.C. concentration (Table III and IV) recorded from both the groups up to 112 days of experimentation ranging from 13.15 to 17.75 millions/mm³, W.B.C. (Table V and VI) ranging from 12.1 to 16.75 thousands/mm³, Haemoglobin (Table VII and VIII) ranging from 7.2 to 10.6 g./100 ml. and Plasma Protein (Table IX and X) ranging from 7.0 to 8.8 g./100 ml and lies within the normal range. Statistical analy sis of the data reveal no significant difference within the groups indicating that supplementation of calcium to the extent of 1.22 %, phosphorus to the extent of 0.63 % and magnesium to the extent of 1.2% seems to have no influence on these parameters.

<u>Serum Calcium, Serum inorganic Phosphorus and Serum Magnesium</u>: Average biweekly serum concentration of calcium (Table XI and XII) recorded for the experimental goats maintained on both the groups up to 112 days of experiment ranging from 7.4 to 9.2 mg./100 ml, serum inorg. phosphate (Table XIII and XIV) ranging from 5.0 to 7.5 mg./100 ml. and serum magnesium (Table XV and XVI) ranging from 2.1 to 5.2 mg./ 100 ml. A trend towards higher serum inorganic phosphate concen tration (mg./100 ml) 5.9 to 6.7 and a trend towards higher serum magnesium concentration from 2.6 to 4.0 in animals maintained on

magnesium concentration from 2.6 to 4.0 in animals maintained on ration B supplemented with magnesium to the extent of 1.2 per cent in the ration could be noticed.

<u>Incidence of Wrinary Calculi</u>: Of the two groups of animals main tained on rations A and B respectively, the animals maintained on Ration B exhibited severe crystalluria which forecasts the initia tion of urinary calculi formation in these animals. The animals were then sacrificed after 124 days for the presence of urinary stones in the bladder, kidney and urethra. The animals maintained on Ration A on post mortem exa mination revealed no traces of urinary calculi. Whereas the animals maintained on Ration B showed urinary calculi in the kidney but were not detected any calculi in the bla dder. The tissue changes on histopathological examination reveal varying degree of changes indicating that the ration B supplemented with magnesium to the extent of 1.2 per cent is positively calculo genic.

<u>Histological findings</u>: For histological examination of urinary organs, paraffim sections of kidneys were made at 5 microns thick ness and stained with haematoxylene and eosin. Histologic examina tion disclosed significant difference between the groups of animals in the pattern of the tissues of the kidneys examined, a brief acc ount of the observations made during the course of the histological studies is given below

On Ration A: There were no significant histopathologic. changes in the kidney. Glomeruli were normal in appearance and in distribution. The epithelial lining of the Bowman's capsule were intact. There were no necrosis or desquation of epithelial lining of the proximal and distal convoluted tubules and ascending and descending limbs of Henle. Tubular casts or other proteinacious deposits were not observed in any part of the renal tubules or collecting ducts. There was no evidence of mineralisation in the kidney tissues. The lumen of the tubule contained homogenous light pink material

<u>On Ration B</u>: There were severe diffuse distruction of the tubules, numerous focal areas of haemorrhage and engargement of inter tubular cappillaries. Many of the tubules showing loss of lining epithelia filled with eosinophilic grannular casts. The glo merular cells showed ballooning, almost filling up the Bowman's capsule and ocassional one showing haemorrhage.

From the above histological observations, it was evident that animals maintained on Ration B manifest severe tissue changes. Ration B shown to be positively calculogenic for goats during the present series of feeding trial and hence this ration was therefore chosen as the basal ration for the next series of feeding trial designed to determine the beneficial role if any of supplemental ammonium chloride and horse gram extract

E. EFFICACY OF AMMONIUM CHLORIDE AND HORSE GRAM EXTRACT ON THE AMELEORATION OF UROLITHIASIS IN GOATS

Experimental Animals: Eighteen male Malabari goats 9 - 12 months of age weighing on an average 19 Kg. after being dewormed and sprayed against ectoparasites were used for the investigation. They were distributed randomly to three experimental groups of six animals each (Groups I, II and III) as uniformly as possible with regard to age and body weight. All the animals were maintained on identical conditions of management and were fed individually. Wholsome water was made available at all times. The experimental duration was 84 days excluding initial and final metabolism trial

Experimental Ration : The concentrate mixture was prepared by using the following conventional feed ingredients

Ingredients	Parts
Wheat	30
Bengal gram	30
Gingelly Oil cake	30
Black gram husk	8
Salt	0.5
Mineral mixture	1.5
Total	100 .0

Throughout the experimental period a roughage concentrate ratio of 1:4 was maintained

The percentage chemical composition (on dry matter basis) of the compounded feed and grass, on proximate analysis are as shown below:

<u>Nutrients</u>	<u>Concentrate</u>	<u>Roughage</u>
Dry matter	93 • 17	34.93
Crude protein	22.94	9.75
Crude fibre	10.01	43.69
Ether extract	4, 12	3.07
Nitrogen Free Extract.	51.66.	39.64
Total Ash	11.27	3.85
Calcium	0.84	0.14
Phosphorus	0.51	0.18
Magnesium	0.31	0.23

The calcium and phosphorus levels have been raised to an estimated level of 1.194 per cent and 0.578 per cent respectively by adding calcium oxide and P_2 O_5 to the above ration

Ration Treatments

<u>Ration A</u>: The basal concentrate ration known to be calculogenic was computed by using the concentrate mixture added with magnesium oxide to raise the magnesium to the estimated level of 1.202 per cent <u>Ration B</u>: Frepared by using basal concentrate ration A fortified with Ammonium chloride to the extent of 1 per cent

<u>Ration C</u>: Was prepared by feeding the basal concentrate, ration A along with Horse gram extract (<u>Dolichos biflorus</u>) to the extent of 1 litre per animal per day in place of drinking water

Every day the horse gram. extract (decoction) was prepared by boiling 1.5 Kg. horse gram with six litre of drinking water (Horse gram and water in the ratio of 1:4) for 60 minutes and then allowed to cool over night. Next day morning the extract was strained and given to animals in Ration C at the rate of one litre per animal per day.

Three experimental groups (I, II and III) of six animals each were fed with the rations A,B and C respectively. Individual feed allowances for each animal for ma intenance was calculated based on the body weight.

The goats were weighed at weekly intervals. Daily feed consumption records were maintained throughout the experimental period. Digestion cum balance trial were carried out initially and at monthly intervals till the termination of feeding trial, quantitative and sep arate collection of urine and feces were carried out with a collection period extended for five consecutive days, using metabolism cages.

The feed samples collected during metabolism trial were subjected to proximate analysis as per standard procedure (AOAC, 1990). Protein in feed, feces and urine were analysed using Kjeltec - 2000 digestion and distillation unit. Calcium and magnesium content of feed, feces and urine samples were estimated by using Atomic Absor ption Spectrophotometer (Perkin Elmer, Model 3110). Urine Phosphorus was determined colorimetrically with Spectrnic - 20 (Milton Roy Co., U.S.A) using commercial phosphorus kit (Qualigens Diagnostics)

Eeed and fecal phosphorus were estimated colorimetrically by nitric acid vanado - molybdate method (AOAC, 1990)

Fresh urine samples were collected at monthly intervals to determine the pH and examined microscopically for the presence of crystals. The crystals were collected and subjected to qualitative chemical analysis (Spot. test. by Winer, 1959)

<u>Haematological studies</u>: Blood samples were collected at monthly intervals by jugular puncture into sterile citrated tubes and in clean dry tubes for serum separation.

Total Erythrocyte count was made by using improved Neubauer counting chamber with 1:200 dilution of blood using Hayem's fluid as diluent

Total Leucocyte Cell Count were made by using Truck's Fluid as the diluent with 1:20 dilution

Haemoglobin content was determined by acid hematin method using haemometer with permanent coloured glass comparison standard.

Plasma protein values were determined colorimetrically by Biuret method using total protein kit supplied by Stangen Immuno diagnostics

Serum calcium and magnesium were estimated by Atomic Absor ption Spectrophotometer (Perkin Elmer Model 3110). Serum Inorganic phosphorus was estimated colorimetrically (Modified metol method) by using commercial phosphorus kit (Qualigen diagnostics)

<u>Slaughter Study</u>: At the termination of the experiment, all the animals were slaughtered and the uninary organs were collected and subjected to detailed examination for the presence of gross calculi and other gross lesions. The calculus materials were collected and subjected to spot tests (Winer, 1959) for identification of its che mical composition

<u>Histopathological Study</u>: The specimens from uninary organs viz., kidney, uneter and bladder were collected and preserved in neutral buffered formalin (10 per cent) for histopathological study. These tissues were processed with routine paraffin embedding method and sections were stained with haematoxylene and Eosin.

<u>Criteria For Evaluation</u>: The criteria used for evaluation of the rations (rations A, B and C) were body weight gain, dry matter consumption, feed efficiency, protein efficiency, haematological parameters, data gathered from digestion cum balance trials, examination of urine, autopsy and histopathological examination.

Results and Discussion

Body weight gain, Dry matter consumption, Feed efficiency and Frotein efficiency values:

Goats maintained on groups I, II and III fed on basal calculogenic ration (Ration A), calculogenic ration supplemented with ammonium chloride (Ration B) and calculogenic ration supple mented with horse gram extract: (Ration C), recorded an average cumu lative weight gain and average daily gain during the experimental period as 5.20, 5.29 and 5.17 Kg. and 61.90, 62.98 and 61.55 g. per day (Tables 1 and 2) respectively, the values being not stat istically significant between the groups indicating that neither supplementation of ammonium chloride nor horse gram extract had any significant influence on the body weight gain of goats, however, group IL had a better weight gain than the animals maintained under groups I and III.

The dry matter consumption recorded for the three groups I, II and III were 928.57, 917.98 and 901.38 g. per day respectively, had not shown any statistical significance between the three groups (Tables 3 and 8). Eeed efficiency (Tables 4 and 8) and protein efficiency (Tables 5 and 8) values for the animals maintained on rations A, B and C being recorded as 15.02 and 2.69, 14.58 and 2.60 and 14.69 and 2.62 respectively, did not exhibit any significant variation among the three groups, indicating that neither supple mentation of ammonium chloride nor horse gram extract had any sig nificant influence on the dry matter intake, feed efficiency and protein efficiency but group II fed supplemental ammonium chloride had a tendancy towards better feed efficiency and protein efficiency than groups I and III

·- 13 -

Haematological parameters such as TEC (Table 9), TLC (Table 10), Haemoglobin (Table 11) and total protein (Table 12) obtained from three groups at monthly intervals, during the exper imental period had no significant difference between the groups ind icating that neither supplemental ammonium chloride nor horse gram extract had any significant influence on these haematological para meters (Table 13). The values recorded were within the normal range throughout the course of experiment showing that the animals were in normal nutritional status

Serum Calcium. Inorganic Phosphorus and Magnesium concentrations

Average serum calcium values (mg./dl.) recorded for the three groups being 8.67, 9.29 and 8.99 respectively for the groups I, II and III. There was significant reduction in serum calcium concentration in group I receiving Ration A when compared to that of groups II and III receiving Rations B and C, whereas no significant difference exhibited between the groups II and III. The results in dicated that high dietary magnesium interfered with the absorption of calcium but supplementation of ammonium chloride and horsegram extract significantly counteracted the influence of high dietary magnesium over calcium absorption in that descending order and maintained the serum calcium concentration within the normal range (Table 14 and 17)

Serum Inorganic phosphorus values (mg./dl.) obtained during the course of the experiment being 6.47, 5.69 and 6.08 for the animals maintained on groups I, II and III, indicating that high dietary mag nesium enhances the absorption of dietary phosphorus, but supplemental ammonium chloride or horsegram extract interfers with the absorption of phosphorus or counteracts the action of high dietary magnesium on phosphorus absorption and maintained the serum inorganic phosphorus concentration within the normal range (Tables 15 and 17).

Average serum magnesium concentration (mg./dl) obtained from the three groups being 4.78, 3.70 and 4.22 respectively for the groups I, II and III, exhibited significant difference among the three groups, indicating that increasing the dietary magnesium causes elevation in serum magnesium due to increased magnesium absorption, whereas supplemental ammonium chloride and horse gram extract had profound influence on reducing the serum magnesium concentration in the respective groups, but the effect was more pronounced by supple mental ammonium chloride than horse gram extract (Tables 16 and 17).

Mineral and Nitrogen balance

Data gathered from mineral and nitrogen balance studies are presented in Tables 18 to 35. Urine Values

<u>Calcium</u>: Urine calcium values recorded for the three groups I, II and III at the beginning and at the end of experimental period were 0.104 and 0.191, 0.109 and 0.120 and 0.108 and 0.151 g./day respectively. Statistical analysis disclosed that goats maintained under group I excreted significantly high urine calcium compared to groups II and III, while no significant difference between the groups II and III, indicating that high dietary magnesium interferes with the utilisation of whereas supplemental ammonium chloride and horse gram extract had counteracted the influence of high dietary magnes ium over calcium utilisation.

Phosphorus: Urine phosphorus values (g./day) obtained from three experimental groups I, II and IIL at the beginning and at the end of the experiment being 0.182 and 0.530, 0.197 and 0.230 and 0.199 and 0.275 g./day respectively. Statistical analysis disclosed that there was significant rise in urine phosphorus excretion in group I when compared to that of animals maintained on groups II and III, indicating that high dietary magnesium interferes with the uti lisation of phosphorus, however, no significant difference was dis cernible between the groups II and III, indicating that supplemental ammonium chloride significantly reduce the urine phosphorus excretion and increase its utilisation whereas the effect is less pronounced in goats maintained on supplemental horse gram extract. <u>Magnesium</u>: Data on urine magnesium (g./day) gathered at the beginning and at the end of the experiment being 0.220 and 1.617, 0.190 and 1.580 and 0.212 and 1.159 g./day for the animals main tained on groups I, IL and III respectively, indicating that no significant variation exists between the groups with regard to magnesium excretion through urine. These results indicated that high dietary magnesium causes increased excretion of this element through urine. Neither supplementation of ammonium chloride nor horse gram extract had any significant influence on the utilisation of magnesium

<u>Nitrogen:</u> Urinary nitrogen excretion (g./day) at the beginning and at the termination of the experiment being 6.904 and 11.445, 6.319 and 10.953 and 6.854 and 10.947 from goats maintained on groups I, II and III respectively, had not shown any significant difference between the groups indicating that neither supplemen tation of ammonium chloride nor horse gram extract had any signi ficant influence on nitrogen utilisation

Faecal Values

Data on faecal calcium, phosphorus, magnesium and nitrogen obtained from the three groups I, II and III at the termination of the experiment being 5.112, 4.304 and 4.472 g./day for calcium, 3.700, 3.530 and 3.613 g./day for phosphorus, 6.101, 6.503 and 5.973 g./day for magnesium and 11.610, 11.997 and 11.665 g./day for nitrogen respectively. Statistical analysis of the data reveal no significant difference between the three groups with regard to the faecal excretion of calcium, phosphorus, magnesium and nitrogen indicating that neither supplementation of ammonium chloride or horse gram extract when fed along with high magnesium calculogenic ration had any significant influence on digestibility of dietary calcium, phosphorus, magnesium and nitrogen

Fercent Retention

<u>Calcium</u>: Average percent retention of calcium in goats main tained on groups I, II and III at the beginning and at the end of the experiment being 62.05 and 59.85, 62.71 and 66.55 and 61.04 and 62.65 per cent respectively indicating that high dietary mag nesium in the diet interferes with the percent retention of calcium There was significant rise in calcium balance during the progress of experiment in group II when compared to that of groups I and III and no significant difference between the groups I and III, indicating that supplemental ammonium chloride enhance the reten tion of calcium, whereas supplemental horse gram extract had not shown any significant influence on percent retention of calcium, but had a trend towards better percent retention of calcium

<u>Phosphorus</u>: Data gathered on percent retention of phosphorus from three groups, I, II and III at the beginning and at the end of the experiment being 46.64 and 35.24, 45.28 and 43.24 and 47.48 and 37.72 per cent respectively, reveal a significantly higher phosphorus balance during the progress of experiment in group II when compared to that of groups I and III, indicating that high dietary magnesium interferes with the percent retention of phos phorus while supplemental ammonium chloride enhances the retention of phosphorus. Supplemental horse gram extract showed only a tendancy to enhance the retention of phosphorus.

<u>Magnesium</u>: Data on percent retention of magnesium obtained from the three groups I, II and III during the beginning and at the end of the experiment being 32.20 and 42.49, 31.85 and 40.01 and 34.00 and 41.80 respectively reveal no significant difference between the groups, indicating that, higher the dietary magnesium, higher will be the percent retention of magnesium. Neither supple mentation of ammonium chloride nor horse gram extract had any sig nificant influence on percent retention of magnesium when fed along with high magnesium calculogenic ration. Percent retention of magnesium values in all the three groups have been increased tow ards the termination of the experimental period, but the increase in magnesium retention was more pronounced in groups I, III and II in that descending order.

<u>Nitrogen</u>: Percent retention of nitrogen recorded for the three groups during the beginning and at the termination of the experiment being 32.60 and 41.30, 34.35 and 43.50 and 31.43 and 40.65 per cent for the groups I, II and III respectively, had no no significant difference between the three groups with regard to percent retention of nitrogen, indicating that high dietary magne sium alone or in combination with either supplemental ammonium chl oride or horse gram extract had any significant influence on percent retention of nitrogen. The percent nitrogen retention was increased in all the three groups, the extent of percent nitrogen retention is better in group II, I and III in that descending order.

Microscopic Examination of Urine.

Microscopic examination of urine for the presence of crystalluria reveal that, crystalluria was absent in group II whereas group III had comparatively less intensity of crystalluria than group I, indicating that supplemental ammonium chloride preventing the crystalluria but not by supplemental horse gram extract. The urine pH recorded during the experimental period for the groups I, II and III being 8.4, 8.0 and 8.3 respectively indicating that both the dietary treatments had not exerted any profound influence on urine pH. The average urine volume recorded for the three groups being 1412.9, 1866.93 and 1774.74 ml./day respectively for the groups I, II and III indicating the poss ible diuretic effect of both the dietary treatments (rations B and C). <u>Postmortem Studies</u>

On postmortem and histopathological examination, group I exhi bited severe gross and histopathological tissue changes in the kidney and bladder and also found to have numerous gross and microcalculi in the kidney, which confirmed the calculogenic effect of Ration A. Goats (group II) fed on ration B had not shown any gross lesions, had only mild histopathological changes in the kidney and bladder and had not found to contain any gross and microcalculi in the kidney, which ind icated the possible beneficial effect of ammonium chloride in the pre vention of urinary calculi, whereas goats (group III) fed on ration C were found to contain less number of gross and microcalculi in the kidney along with gross and histopathological changes in group I, which indicate that horsegram extract is not effective in the preven tion of urinary calculi completely, but had a tendancy to reduce the incidence of urinary calculi when supplemented along with high mag nesium calculogenic ration. <u>Conclusion</u>: From an overall assessment of the data gathered during the course of feeding trial, it can be reasonably concluded that a ration having 1.194 per cent calcium, 0.578 per cent phosphorus and 1.202 per cent magnesium was found to be calculogenic for goats. Supplementation of ammonium chloride along with high magnesium cal culogenic ration at the rate of one per cent level prevented the calculiformation in goats, while horse graf extract had not preven ted the calculi formation completely but had a tendancy to reduce the occurrence of calculi

C. <u>EFFECT OF SUPPLEMENTATION OF ELEMENTAL SULPHUR, CALCIUM OXIDE</u> AND VITAMIN D ON THE AMELEORATION OF URINARY CALCULI IN GOATS

Twenty four Malabari male goats in the age group of 10 to 14 months were utilised for the study. They were distributed randomly into 4 groups of 6 animals each (Groups I, II, III and IV) as unifo rmly as possible with regard to age and weight. All the animals were maintained on identical conditions of management and fed individually. Wholesome water was provided <u>ad</u>. <u>libitum</u>. Guinea grass was fed as roughage. Experimental duration is 120 days.

Ration Treatment

Ration A: Control calculogenic ration containing calcium to the extent of 1.194 per cent, phosphorus 0.578 per cent and magnesium 1.202 per cent

<u>Ration B:</u> Ration A supplemented with elemental sulphur to the extent of 1.044 per cent

<u>Ration C</u>: Ration A supplemented with calcium to the extent of 2.2 per cent

Ration D: Ration A supplemented with Vitamin D_3 to the extent. of 550 I.U. /Kg. feed

All the groups of animals are assigned to each ration and the experimental duration is 120 days.

The experiment is in progress

CALCULOGENESIS IN ALBINO RATS

Numerous studies have been reported in the literature on the effect of the mineral content of diets on the severity of dental caries in rats. In the course of similar investigations Losee and Gerende (1957) observed that numerous rats died on some diets which appeared which appeared to be adequate. Autopsies revealed extreme distension of bladder, accompanied by extensive stone formation in the urinary apparatus (Vanreen, Lyon and Losee, 1958).

Investigations on urinary calculi in experimental animals have led to valuable information concerning the factors which are predisposing to this condition. Osborne, Mendel and Ferry (1917) demonstrated the influence of vitamin A deficiency on stone formation while Rosenow and Meisser (1922) clearly illustrated the effect of chronic infection. Keyser (1945) and Wilson, Benjamin and Leahy (1945) directed attention to high dietary minerals in causing urinary calculi while Schneider and Steenbock (1940), indicated that diets extremely low in phosphate would lead to urolithiasis. Since Albino Rats rarely produce calculi under normal conditions, some method of inducing calculogenesis is necessary before urolithiasis can be studied.

During the course of an investigation in albino rats attempts have been made to induce urinary calculi by fortifying the basal diet (Ration A) with magnesium to the extent of 1 per cent (Ration B), magnesium plus ammonium oxalate to the extent of 0.5 per cent each respectively (Ration C), ammonium oxalate to the extent of 1 per cent (Ration D), calcium to the extent of 1.6 per cent (Ration E), urea to the extent of 1 per cent (Ration F) and urea plus ammonium oxalate 0.5 per cent each respectively (Ration G), in order to probe in depth whether these treatments will induce urinary calculi in albino rats.

<u>Experimental</u>

Weanling male albino rats of the Sprague - Dawley strain were used in this investigation. At 30 days of age , rats weighing between 30 and 45 g. were randomly distributed into the desired groups of 6 animals each and placed in individual polypropylene cages. Food and water were provided <u>ad libitum</u>.

All the experimental animals were weighed weekly and the average body weights in each group calculated. Feed consumption data was recorded daily

To follow any histological and metabolic changes that might occur during the development of urolithiasis, the rats were sacrificed after three months. Blood and urine samples were taken for mineral analysis and the kidney and bladder were examined for microcalculi. The kidney tissue was collected and preserved in neutral buffered formalin for histopathological examination. These tissues were pro cessed with routine paraffin embedding method and sections were stained with haematoxylene and eosin.

Frior to sacrifice 7 day mineral metabolism trials were conducted. Representative samples of both feces and urine were taken separately and were stored in deep freezer. The samples collected from each rat were preserved, during the entire collection period and later pooled and used for further analysis

The feed samples collected during metabolism trial were subjected to proximate analysis as per standard procedure (AOAC, 1990). Nitrogen in feed and feces and urine were analysed using Kjeltec 2000 digestion and distillation unit

Calcium and magnesium content in feed, feces and urine samples were estimated by using Atomic Absorption Spectrophotometer (Ferkin Elmer, Model 3110). Urine phosphorus was determined colori metrically with Spectronic 20 (Milton Roy Co., USA) using commercial phosphorus kit (Qualigens Diagnostics).

Feed and fecal phosphorus were estimated colorimetrically by nitric acid vanado - molybdate method (AOAC, 1990).

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<u>Diet:</u> Ingredient composition is presented in Table 1

<u>Ingredients</u>	<u>Parts</u>
Gingelly oil cake	30
Bengal gram	30
Wheat	30
Black gram husk	. 8
Mineral mixture	1.5
Salt	0.5
	100.0

Experimental outline with treatment variables are shown in Table 2

		Treatme	ent var	iable			:,
Variables	· · · · · · · · ·			<u></u>	<u></u>		
	A 6	в 6	С 6 -	D 6	E . 6	₽ • 6	G . 6
Calcium	0.81	0.81	0.81	0.81	1,6	0.81	0.81
Pho spho ru s	0.44	0 -4 4	0.44	0.44	0.44	0.44	0.44
^M agnesium	0.31	1.00	0.50	0.31	0.31	0.31	0.31
Amm. Oxalat	е -	_	0.50	1.00		-	0.50
Urea	-	_	_ ·		_	1.00	0.50

<u>Metabolism Studies</u>: Calcium, phosphorus and magnesium metabolism studies have been carried out using the experimental rats under different dietary treatments. The results are shown under separate heads

<u>Calcium</u>: Out of the 7 dietary treatments, animals maintained on ration E fortified with calcium to the extent of 1.6 per cent in the ration exhibited slight increase in serum calcium level (Table 3) when compared to other groups. Naturally the calcium concentration in urine and feces, with a higher percentage retention of the mineral could be noticed. In group E, supplemental magnesium to the extent of 1 per cent in the ration seems to enhance the utilisation of calcium and as such the per cent retention of calcium is also increased, so also in group D supplemented with ammonium oxalate. <u>Phosphorus</u>: In all the groups phosphorus level in the ration is almost identical (Table 4). It seems that supplementation of mag nesium interferes with phosphorus utilisation thereby increased excretion of phosphorus through urine and consequent reduction in per cent retention.

Magnesium: Supplementation of magnesium to the extent of 1 per cent causes increased urinary excretion of magnesium through urine in animals maintained on ration B and C (Table 5). But supplementa tion of calcium to the extent of 1.6 per cent seems to decrease the urinary excretion of magnesium through urine and consequent incre ase in retention of magnesium

<u>Incidence of Calculi</u>

Renal lithiasis was manifested in three ways: Froduction of stones, formation of small white flakes detectable with the dis secting microscope and calcification in the renal tubules and in the renal tissues. Only in groups B, C, D and E had an incipience of calculi formation (Table 6). It manifests in the form of white flakes in the pelvis of kidney which do not have the colour, crys talline appearance or hardness of stones. The stones did not appear to interfere with the feed consumption of the experimental animals.

The result of the metabolism study and post mortem and histopathological examination of the tissues of the experimental animals indicate that there is an initiation of calculi formation in high magnesium supplemented groups (groups B and C) whereas in groups D and E supplemental calcium and ammonium oxalate seems to a lesser extent to induce calculi formation in rats (Table 6).

Further experiments on efficacy of citric acid and DL - Methionine in the ameleoration of glycolate induced urinary calculi in growing rats is in progress.

•	;. !		GROUP	4 -			
		B			E		
Body weight of rats (g.)	123.3	123. 2	124.0	125.0	123.7	122.0	122.3
Serum Calcium (mg./100 ml.)	9•45	9.25	9.32	9 . 10	9.85	9.25	. 9.35
Calcium intake (mg./day)	104.0	1 0 4.0	102.0	102.0	200.0	102.0	101.0
Urinary Calcium (mg./day)	15.5	12.5	16.2	14.5	32.5	13.5	12.5
Faecal Calcium (mg./Day)	60.0	58.5	62.5	54.2	90 . 2	61.0	60.5
Apparant Calcium absorption (mg./		45.5	39.5	45.8	109.8	41.0	40.5
Calcium Retentio (mg./day)	n 28.5	33.0	23.3	31 .3	77•3	27.5	28.0
Fercent Retentio (%)	n 27.4	31.73	23.36	31.30	77.30	26 .96	27.7

TABLE 3 Rat Experiment - Effect of Dietary Treatment on Calcium Metabolism

TABLE 4 Rat Experiment - Effect of Dietary Treatment on Phosphorus Metabolism										
	 A	B	GHOUP C		<u> </u>					
Body weight of rats (g.)	1123.3	123.2	124.0	125.0	123-7	122.0	122.3			
Serum Phosphorus (mg./100 ml)	6.25	7.65	7.25	7.15	7.05	6.25	6.25			
Phosphorus int. (mg./day)	65. 6	66.1	65.5	66.6	62.6	63.9	67.3			
Urinary Phosph. (ng./day)	19.2	25. 2	15.8	18.6	12•5	17.5	15.8			
Faecal Phos. (mg./day)	31.2	32.4	31.2	30.8	31.6	2 9. 0	3 5.1			
Apparant Phos. absorption(mg./d	34•4)	33.7	34.3	35.8	31.0	34.9	32.2			
Phosphorus Retn. (mg./day)	15.2	8.5	18.5	17.2	18.5	17.4	1.6.4			
Percent Retn. (%)	23.17	12.86	28 .24	25.82	29.55	27.23	24.37			

TABLE 5

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Rat Experiment - Effect of Dietary Treatment on Magnesium

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Metabolism

					~ _ ~		
		GR	OUP				
	A	- В	C,	D	ਜ਼	P	G
Body weight of trats (g.)	123-3	123.2	124.0	125.0	123.7	122.0	122.3
Serum Magnesium (mg./100 ml.)	2.75	4.85	4.25	3.25	315	3.02	3.04,
Magnesium intake (mg./day)	45.9	147.8	74.4	46.5	46.0	45.4	45.5
Urinary Magnesium (mg./day)	¤ 9 . 5	30.2	22 .5	9.2	8.5	9.5	10.4
Faecal Magnesium (mg./day)	13 -9	40.4	16.7	16.8	9.1	13.4	11.3
Apparant Magnes- ium abs. (mg./d)	32.0	107.4	57.7	29.7	36.9	32.0	34.2
Nagnesium Ret. (mg./day)	22.5	77.2	35.2	20.5	28.4	22.5	23,8
Fercent Retn. (%)	49.0	52.2	47.3	44 . 1	61,.7	49.6	52 . 3

TABLE 6

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Rat Experiment - Effect of Treatment on incidence of Renal Calculi in Rats

GROUP	No. of Rats	Norm.	Hist.	Fl	St
			·		
A	6	6	-		
В	6	_	6	3	
0	6	-	6	3	-
D	6	_	6	_	-
Ξ	6	-	6	_	
F	6	-	6	_	-
G	6	-	6		-
Norm.		 animal wit ithiasis	h no gross or	histologica	
Hist.		owing only r al damage		evidence of	nephrocalcinosis,
Fl.	Rats hav	ving white	flakes but no	stones in	the kidney
St.	Rats sho microsco		al stone forma	ation detect	able with

- 16. Summary of the trial : Experiments pertaining to calculogenesis and ameleoration in both ruminant and monogastric animals is in progress.
- 17. Results which can be exploited on pilot/field scale : Nil
- 18. Fublication : Nil
- 19. Contribution made by the cooperators Well cooperated
- 20. SIGNATURE

C.S. JAMES

Name.

Designation:

Professor & Principal Investigator

Date : 1.5.1999

DIRECTOR IN CHARGE/STATION

RCH versity 880654



Animal :		• • •						DAYS			· · · · · ·					·	
No:	9]	7	14	21	28	35	42	49	56	63	70	77	84	91	98	105	112
1.	23.9	26.0	25.5	25.7	28.5	28.1	29.3	28.8	28.8	29.2	29.5	30.1	30.5	30.9	31.2	31.5	3 2.3
2.	21.0	22.0	22.3	22.7	25.5	25.6	26.3	26.4	27.2	27.5	27.8	28.2	28.5	28.8	2 9.2	2 9 .5	30.0
3.	15.4	17.5	17.5	18.4	21.0	21.2	22.0	22.3	23.1	23.5	23.8	24.1	24.5	24.8	25.2	25.5	25.
4.	: 14.0 §	15.5	15.4	17.0	18.4	21.6	19.8	20.9 ,	21.3	21.6	22.0	22.3	22.5	22.8	23.1	23.5	23.
5.	17.0	17.3	18.7	18.8	21.1	21.8	23.0	23.1	24.0	24.2	[•] 24.5	24.9	25.2	25.5	25.8	26.1	26.
6.	14.9	14.0	14.2	15.3	15.8	16.7	16.8	17.3	18.8	19.0	19.2	19.5	19.8	20.1	20.5	20.8	21.
7.	13.8	14.8	15.5	16.0	17.2	17.8	18.8	19.8	20.4	20.8	21.2	21.5	,21.8	22.1	22.4	22.7	23.
8.	16.3	15.5	16.0	17.2	18.2	18.8	19.8	20.4	20.8	21.0	21.4	21.7	22.0	22.1	22.5	22. 7	23.
9.	24.0	24.2	24.5	25.5	27.6	. 27.7	27.8	28.3	29.3	29.8	30.0	30.5	30.8	31.2	31.5	31.8	32.
		1 V.					l										
Average	17.82	18.5	18.9	19.6	21.5	22.1	22.8	23.0	23.7	24.1	24.4	24.7	25.1	25.4	25.7	26.0	26.

TABLE I • <u>BODY WEIGHT GAIN' IN' KG OF ANIMALS MAINTAINED ON RATION A</u>

TABLE I	
BODY WEIGHT GAIN' IN Kg OF ANIMALS MAINTAINED ON RATION	в

,	. <u></u>						• • ·	· · · · · ·		 			·				
Ànimal						•	. •	DAYS		· · ·							
No:	0	. 7	14	21	28	35	42	49	56	63	70	77	84	91	98	105	112
10.	24.0	24.0	24.5	25.5	27.6	26.6	27.8	28.3	29.3	29.5	29.8	30.0	30.4	30.7	30.9	31.2	31.8
11.	22.0	23.0	24.8	24.Ż	26 <i>.</i> 4	26.9	28.0	28.1	28.8	30.1	30.5	30.8	31.0	31.5	31.8	31.9	32.0
12.	12.2	12.5	12.5	13.6	15.0	14.9	15.2	1 6 .1	17.0	17.2	17.5	17.8	17.9	18.0	18.5	18.7	[,] 19.0
13.	13.8	14.5	15.5	16.0	17.2	21.0	18.8	19.8	20.4	20.8	20.9	21.2	21.5	21.8	22.1	22.2	22.9
14.	16.3	15.5	15.5	16.5	18.4	19.3	20.2	20 .3	21.3	21.8	22.0	22.2	22.5	22.8	23.1	23.3	23.5
15.	18.0	19.0	20.0	21.3	23.1	24.6	23.4	25.1	24.8	25.2	25.5	25.8	26.2	26.8	27.1	27.2	27.5
16.	22.0	23.0	24.8	24.2	26.4	26.9	28.0	28.1	28.8	29.0	29 .2	29.5	30.1	30.4	30.7	31.0	31.5
17.	12.2	12.5	12.8	13.6	14.2	14.9	15.2	16.1	17.0	17.2	17.5	17.8	18.2	18.5	18.9	19.4	19.7
18.	18.0	19.0	20.0	21.3	22.1	21.5	22.0	22.5	22.8	23.0	23.5	23.8	23.9	24.2	24.5	25.8	26.1
			l						, -								
Average	17.6	18.1	18.9	19.6	21.2	21.8	22.1	22.7	23.4	23.8	24.0	24.3	24.6	25.0	25.3	25.6	26.0

Anitnal									
No.	0	14	28	42	56	70	84	98	112
, 1	17.80	17.55	17.45	17.85	17.25	17.28	17.55	17.25	17.35
2.	16.59	16.55	16.75	16.25	. 16.85	16.80	16.55	16.75	16.45
3.	14.7 9	14.58	14.75	14.45 .	14.85	14.58	14.75	14.95	14.79
4.	16.8	16.45	16.75	16.45	16.55	16.45	16.85	16.75	16.44
5.	16.79	16.65	16.85	16.52	16.42	16.45	16.55	16.54	16.75
6.	15.06	15.42	14.95	14.98	15.15	15.25	15.15	+ 15.05	14.95
7	16.35	16.35	16.25	16.28	16.18	16.25	16.30	16.35	16.35
8.	16.44	16.42	16.21	16.52	16.12	16.45	16.25	16.45	16.52
9.	16.35	17.25	17.52	17.28	17.55	17.15	17.25	17.32	17.25
È				1					
Average	16.44	16.37	16.39	16.29	16.32	16.30	16.34	16.38	16.32

RED CELL CONCENTRATION (Millions/mm³) OF THE ANIMALS MAINTAINED ON RATION A

TABLE III

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	<u>ה</u> .	UL	، نگ	ΤV	

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RED CELL CONCENTRATION (Million 1/1nm3) OF THE ANIMALS MAINTAINED ON RATION B

ⁱ Animal			1.1		DAYS		•	·	
No.	0	14	28	.42	56	70	84	98	112
. 10.	16.58	16.75	16.25	16.48	16.45	16.55	16.45	16.75	16.35
11.	15.12	15.15	15.25	15.14	15.15	15.25	15.25	15.15	15.35
12.	17:80	17.52	17.68	17.25	17.45	17.25	17.35	17.80	17.75
13.	17.97	17.85	17.95	17.58	17.62	17.85	17.55	17.65	17.75
14.	13.16	13.15 [;]	13.75	13.25	13.45	13.25	13.45	13.35	13.65
15.	16.47 •	16.75	16.45	16.25	16.38	16.45	16.55	16.25	16.75
. 16.	15.08	15.25	15.18	15.25	15.15	15.15	1,5.25	15.15	15.25
17.	16.85	16.52	,16 . 85`	16.75	16.52	. 16.75	16.25	16.45	16.25
18.	17.25	17.15	16.95	17.15	17.22	17.15	17.25	16.95	17.25
-									
Average	16.25	16.23	16.27	16.12	16.15	16,18	.16.15	16.17	16.26
						<u>_</u>			

Animal					DAYS	· · · ·			
No.	0	14	28	42	56	70	84	98	112
· 1	15.00	15.25	15.25	15.15	15.25	15.25	15.15	15.3:	.15.25
2.	16.70	16.25	15.95	15.95	16.15	16.25	16.55	16.45	16.65
3.	13.75	13.85	14.25	1 3.9 5	. 14.15	13.85	14.25 .	14.15	14.05
4.	13.50	- 13.25	13.45	13.25	13.55	13.25	13.55	13.45	13.35
ʻ5 .	12.60	12.55	12.25	12.54	12.48	12.25	12.65	12.35	12.45
6.	16.05	15.95	16.15	16.25	16.15	16.25	16.05	16.15	16.25
7.	14.50	14.25	14.28	14.35	14.50	14.25	14.15	14.28	14.32
8.	12.10	12.12	12.50	12.22	12.25	12.12	12.28	12.15	12.25
9.	16.85	16.15	16.25	16.28	16.32	16.75	16.52	16.55	· ·16.15
• •									
Average	14.56	14.40	14.48	14.44	14.53	14.47	14.57	14.54	14.52

WBC CONCENTRATION (Thousan 1: /mm³) OF THE ANIMALS MAINTAINED ON RATION A

TABLE V

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TABLE VI

WBC CONCENTRATION(Thou sand/mm³) OF THE ANIMALS MAINTAINED ON RATION B

Animal				•	DAYS			, i .	•
No.	0	14	28	42	56.	70	84	98	112
10.	14.00	15.25	14.85	14.65	15.15	14.20	14.55	14.25	14.15
11.	15.85	15.55	15.65	15.72	15.80	15.75	15.80	15.7.	15.55
12.	12.65	13.25	12.55	• 14.15	13.65	12.75	13.25	13.45	13 .3 5
13.	12.75	12.72	12.55'	12.35	12.75	12.72	12.75	13.15	13.25
14.	13.32	13.52	13.25	13.45	13.15	13.35	13.15	13.25	13.45
15.	14.55	14.25	14.55	14.85	14.65	14.50	14.55	14.25	. 14.30
16.	15.45	15.25	15.65	15.45	· 15.25	15.50	15.45	15.25	15.25
17.	15.85	15.65	15.75	15.55	15.65	15.45	. 15.85	15.35	15.55
18.	14.6	14.75	14.55	14.85	14.75	14.25	14.65	14.75 •	.14.55
				1 1 1					
Average	14.35	14.47	14.37	14.56	14.56	14.27	14.44	14.38	14.38

TABLE VIII

HAEMOGLOBIN CONCENTRATION(Gram%) OF THE ANIMALS MAINTAINED ON RATION B

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Animal				_	DAYS		<u> </u>	, ,	
No.	0	14	28	42	56	70 .	84	98	112
10.	9.2	9.0	8.5	8.9	9.2	9.2	9.1	9.2	9.0
11.	7.6	7.5	8.0	8.5	8.5	8.4	8.5	8.2	8.0
12.	8.4	8.5	8.7	8.6	8.8	8.4	8.5	8.2	8.5
13.	8.8	9.0	8.5	8.8	8.5	8.5	8.8	9.0	8.8
14.	9.4	9.2	9.0	9.5	9.2	9.0	9:1 ·	[;] 9.0	8.9
15.	7.6	7.8	8.0	.8.0	7.8	7.5	7.2	7.8	7.6
16.	10.2	9.5	9.8	9.5	9.8	9.2	9.2	9.4	9.5
17.	7.8	7.9	8.0	8.5	8.0	8.2	. 8.4	8.0	8.2
18.	9.2	9.5	9.0	8.9	9.2	9.0	8.8	9.2	8.8
Average	8.7	8.7	8.6	8.8	8.8	8.6	8.6	8.7	8.6

TABLE VIII

HAEMOGLOBIN CONCENTRATION(Gram%) OF THE ANIMALS MAINTAINED ON BATION B

-

Animal		•			DAYS	. :			<u> </u>
No.	0	14	28	42	56	70	84	98	112
10.	9.2	9.0	8.5	8.9	9.2	9.2	9.1	9.2	
11.	7.6 ·	7.5	8.0	8.5	8.5	8.4	8.5	8.2	8.0
12.	8.4	8.5	8.7	8.6	8.8	8.4	8.5	8.2	8.5
13.	8.8	9.0	8.5	8.8	8.5	8.5	8.8	9.0	8.8
14.	9.4	9.2	9.0	9.5	9.2	9.0	9.1	9.0	8.9
15.	7.6	7.8	8.0	8.0	7.8	7.5	7.2	7.8	7.6
16.	10.2	9.5	9.8	9.5	9.8	9.2	9.2	9.4	9.5
17.	7.8	7.9	8.0 i	8.5	8.0	8:2	8.4	8.0	8.2
18.	9.2 ·	9.5	9.0	8.9	9.2	9.0	8.8	9.2	8.8
Average	8.7	8.7	8.6	8.8	8.8	8.6	8.6	8.7	8.6

: '	 :								
Animal					DAYS	· · ·			
No.	• • •	14	28	42	56	70 ·	84	98	112
<u> </u>	8.8	8.8	8.7	7.5	8.5	8.8	8.4	8.6	- 8.8
2.	8.0	8.0	7.5	7.9	8.5	8.2	8.4	8.5	8.6
3.	8.8	8.5	8.7	8.5	8.6	8.4	8.4	8.5	8.6
4.	7.0	7.5	7.2	8.0	8.1	7.9	8.1	8.2	8.0
5.	7.0	7.5	7.2	7.2	8.0	7.5	7.5	7.6	7.5
5	· [:] 8.7	8.5	8.2	8.5	8.6	8.8	8.4	8.2	8.5
7.	7.0	7.5	7.2	7.5	7.7	7.5	7.2	. 7.5	7.5
8.	, 7.0	7.2	7.5	7.7	7.5	7.5	· 7.5	7.6	7.6
9.	8.8	8.5	8.7	[°] 8.5	8.2	8.4	8.2	8.6	8.5
werage	8.0	8.0	7.9	7.9	8.2	.8.1	8.0	8.1	8.2

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TABLE IX

TABLE	Х	
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PI ASMA PROTEIN CONCENTRATION(Gram%) OF THE INMALS MAINTAINED ON RATION B

Anim 1			DAYS					
No. 0	14	28	42	56	70	84	98	112
10. 8.6	8.5	8.4	8.6	8.7	.8.6	8.4	8.2	8.8
11. 8.7	8.6	8.4	8.7	8.8	8.8	8.6	8.4	8.8
12. 7.4	7.5	7.2	7.5	7.7	7.5	7.8	• 7.6	7.4
13. 7.6	7.8	8.0	7.5	7.2	7.5	7.2	7.8	8.0
14. 7.6	7.8	7.5	7.6	7.7	7.8	7.6	7.5	7.8
15. 8.0	8.2	7.8	8.0	8.5	8.0	8.2	8.4	8.2
16. 7.2	7.5	7.5	7.6	7.5	7.5	7.8	7.4	7.6
17. 7.0	7.2	7.5	7.2	7.8	7.2	7.0	7.6	7.8
18, 7.8,	7.5	7.2	7.7	7.8	7.2	7.8	7.6	7.4
					· ·			
Average 7.8	7.8	7.7	7.8	3.0	7.8	7.8	7.8	8.0

TALLE XI

SERUM CALCIUM CONCENTRATION(mg%) DF THE ANIMALS MAINTAINED ON RATION A

				an a			!	' I ' .
Animal			ľ	L AYS				
No.	0	14	28	42 56	70	84	98	112
1	8.5	8.6	8.6	8.3 8.5	8.6	8.9	8.8	8.9
, 2.	8.9	8.6	9.0	8.8	8.6	8.8	8.9	8.3
3.	8.2	8.5	8.4	8.5 8.8	8.5	8.4	8,8	8.8
4.	8.3	8.5	8.5	8.6	8.5	8.4	8.8	9.0
5,	8.8	8.9	9.0	8.8 8.5	8.8	8.9	9.0	9.2
5	7.9	8.5	8.2	8.5 8.2	.8.5	8.2	8.4	8.2
7.	8.0	8.2	8.2	8.5 8.4	8.4	8.0	8.2	8.4
8.	8.4	8.2	8.5	8.6 8.4	8.4	8.2	8.6	8.4
9.	7.5	8.0	8.5	8.0 8.2	7.9	8.5	8.2	8.8
Average	8.3	8.4	8.5	8.5 8.5	8.5	8.5	8.6	8.7

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 TO M DT 17 N/TT
TABLE XII
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SERUM CALCIUM CONCENTRATION (mg%) OF THE ANIMALS MAINTAINED ON RATION B

			· · · ·					<u> </u>
Animal		·	ų ¹ . 1	DAYS	>		• •	
No.	0	14	28	42 56	70	84	98	112
10.	7.6	8.0	8.2	8.0 7.8	7.6	7.4	7.8	8.2
11.	8.6	8.4	8.6	8.8 8.5	8.6	8,4	8.8	8.5
12.	7.8	8.0	. 8.2	8.0 8.5	8.2	7.2	8.0	8.5
13.	8.6	8.8	8.6	8.5 8.8	8.6	8.8	8.6	8.4
14.	7.7	7.5	8.0	8.5 8.8	7.8	8.0	8.2	8.8
15.	8.0	8.4	8.5	8.8 8.5	8.2	8,4 •	8.6	8.6
16.	8.9	8.8	8.8	8.5 8.5	9.0	8.8	8.5	8.6
17.	8.8	9.0	8.8	8.2 8.8	8.6	8.4	8.8	9.0
18.	8.6	8.4	8.2	8.5 8.6	8.6	8.4	8.5	8.6
	,							
Average	8.3	8.4	8.4	8.4 8.5	- 3.4	8.2	8.4	8.6

TABLE XIII

SERUM INORGANIC PHOSPH ATP: CONCENTRATION(mg%) OF THE ANIMALS MAINTAINED ON RATION A

	1.	:								
Animal		<u> </u>			DAYS	· ·	· · · · ·	<u> </u>		
No.	0.	14	28	42	56	70	84	98	112	
1	5.2	5.1	5.3	5.5	5.0	5.2	5.0	5.5	5.3	
2.	5.0	5.2	5.1	5.5	5.2	5.3	5.0	5.3	5.5	
3.	5.7	5.5	5.6	5.4	5.5	5.5	5.2	5.2	5.4	
4.	6.0	6.2	6.1	6.4	6.2	6.3	6.2	6.2	6.3	
5.	5.5	5.3	5.4	5.2	5.5	5.5	5.2	5.3	5.2	
5	7.7	7.5	7.6	7.2	7.5	7.3	7.2	7.3	7.5	
7.	7.5	7.5	7.2	7.2	7.7	7.5	7.2	7.4	7.5	
8. ·	5.2	5.4	5.5	5.6	5.0	5.2	5.4	5.6	5.4	
9.	5.2	5.4	5.5	5.6	5.4	5.4	5.2	5.4	5.2	
			,			, , , , , , , , , , , , , , , , , , ,				
Average	5.9	5.9	5.9	6.0	5.9	5.9	5.7	5.9	5.9	

TABLE XIV

SERUM INORGANIC PHOSPHATE CONCENTRATION(mg%) OF THE ANIMALS MAINTAINED ON KATION B

				;						
Animal		 ;			DAYS					
No.	0	14	28	.42	56	. 70	84	98	112	
10.	4.6	4.3	5.0	5.2	5.6	5.4	5.2	5.4	- 5.5	
11.	6.0	6.5	6.2	6.1	6.5	6.4	6.4	6.5	6.6	
12.	7.0	· 7.1	6.5	6.5	7.1	7.2	· 7.2	7.3	7.2	
13.	6.2	6.5	6.8	7.0	7.1	7.0	7.2	7.1	7.2	
14.	6.7	7.0	7.1	6.9	7.1	7.2	7.2	7.2	7.3	
15.	7.5	7.2	7.4	7.5	7.5	7.2	7.4	7.3	7.5	
16.	3.4	4.2	4.3	4.5	5.0	5.2	5.0	5.0	5.2	
17.	5.2	5.5	5.8	6.0	6.5	6.4	6.5	6.6	6.5	
18.	6.1	6.5	6.4	6.8	7.0	7.0	6.9	6.9	7.1	
Average	5.9	5.1	6.2	6.3	6.6	6.6	6.6	6.6	6.7	

TABLE	xv

SEFUM MAGNESIUM CONCENTRATION(mg%) OF THE ANIMALS MAINTAINED ON RATION A .

Animal	DAYS										
No.	0	. 14	28	42	56	70	84	98	112		
1	2.9	3.2	2.5	2.8	2.9	3.0	. 2.8	2.9	2.9		
2.	2.8	2.5	2.8	3.2	2.8	3.0	3.1	2.8	3.0		
3.	2.5	2.2	2.5	2.8	2.8	2.6	, 2.6	2.4	2.8		
4	2.9	2.2	2.5	2.9	2.9	3.0	2.9	2.9	3.0		
5.	2.9	2.8	3.0	3.0	3.1	2.8	2.9	3.0	3.1		
6	3.6	2.8	2.9	3.0	3.2	3.5	3.4	3.5	3.6		
7.	3.4	3.8	3.1	3.0	2.9	3.5	3.0	3.2	3.0		
8.	2.3	2.4	2.5	2.5	2.8	.2,5	3.0	3.0	2.8		
9.	2.4	2.5	2.8	3.0	3.0	2.8	2.5	3.0	2.7		
		N									
Average	2.8	2.6	2.7	2.9	2.9	3.0	2.9	3.0	3.0		

TABLE XVI

SERUM MAGNESIUM CONCENTRATION(mg%) OF THE ANIMALS MAINTAINED ON RATION B

Animal	•	•			DAYS		• •		
No.	0	, 14	28	42	56	70	.84	98	112
10.	2.1	2.5	2.8	2.5	3.2	3.3	3.2	3.4	3.5
11.	2.3	2.5	2.8	3.0	3.2	3:5	3,4	3.8	3.8
12.	2.2	2.5	2.8	3.2	3.3	3.5	3.5	3.7	3.8
13.	2.9	3.2	3.4	3.8	4:.0	3.8	4.0	4.2	4.1
14.	2.8	3.2	3.5	3.9	4.5	3.8	3.9	4.0	4.1
15.	2.1	2.5	2.5	2.8	3.0	3.2	3.4	3.4	3.5
16.	3.2	3.5	3.9	3.8	4.5	4.4	4.5	4.6	4.5
17.	2.8	3.5	3.8	4.0	4.8	5.0	, 4.8	5.2	5.0
18.	2.2	2.8	3.2	3.5	4.0	4,2	· 4.0	3.8	4.0
	· ·								
Average	2.6	2.9	3.2	3.4	3.8	3.9	3.9	4.0	4.0

Treatments	Ration A	Ration B	Ration C
Number of animals	6)	6	6
• . 0 day	22.39±0.93	22.53 <u>±</u> 1.24	22.30±1.01
l week	22.85±1.04	22.99 <u>+</u> 1.30	22.75±1.04
2	23.29±1.25	23.44 <u>+</u> 1.35	23.20±1.08
3 '	23.73±1.29	23.88±1.35_	23.64 ±1.1 3
4	24.16±1.34	24.32±1.39	24.07±1.17
5	24.58±1.39	24.75±1.43	24.50±1.22
5 <u>.</u>	24.99±1.42	25.18 ±1 .49	24.93±1.24
7.	25.41 <u>±</u> 1.45	25.61±1.55	25.35 <u>+</u> 1.28
3	25.84 ±1. 46	26.05±1.59	25.78±1.31
)	26.28±1.45	26.50 <u>+</u> 1.64	26.20±1.37
.0 .	26.72±1.48	26.95±1.69	26.63±1.41
.1	27.15±1.46	27.39 <u>+</u> 1.73	27.06±1.46
.2	27.59±1.52	27.82±1.79	27.47±1.51

Table 1. Average weekly body weight (kilogram) of animals maintained on the experimental rations (Rations A, B and C)

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Table 2.	Summarised data on average initial body weight, average final body weight, average cumulative weight gain and average daily gain of animals maintained on three dietary regimes (Rations A, B and C)

Treatments	Ration A containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202%	Ration B containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Ammonium chloride-1%	Ration C containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Horse gram extract 1 lit/animal/day
Number of animals	6	6	6
Initial body . weight (kg)	22.39±0. 93	22.53±1.24	22.30±1.01
Final body weight (kg)	27.59±4.52	27.82±1.79	27.47±1.51 ,
Cumulative weight gain 84 days (kg)	5.20±0.429	5.29±0.445	5.17±0.387
Average daily gain (g)	61.90±0.00 4	62.98±0.006	61.55±0.002
r			

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0-7 days	727.64	738.06	720.42
7-14	755.40	479.08	740.78
14-21	781.95	787.82	766.18
21-28	808.92	797.68	790.54
28-35	855.65	856.87	835.87
35-42	916.82	890.26	884.75
42-49	953.56	939.27	920.13
19-56 ⁻	1017.31	998.19	990.56
56-63	1056.37	1033.38	998.12
53-70	1066.53	1052.60	1047.04
70-77	1075.40	1070.59	1078.12
77-84	1127.32	1102.03	1054.00

Table 3. Average daily dry matter intake (g) of the animals maintained on the experimental rations (Rations A, B and C)

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0-7 days	11.07	11.23	11.20
7-14	12.02	11.65	11.52
14-21	12.44	12.53	12.19
21- 2 8	13.17	12.69	12.86
28-35	14.26	13.86	13.60
35-42	15.65	14.49	14.40
42-49	15.89	15.29	15.33
49-56	16.56	15.88	15,96
56-63 .	16.80	16.07	16.63
63-70	16 .9 6	16.37	17.04
70-77	17.50	17.03	17.55
77-84	17.93	17.94	17.99

Table 4. Average weekly feed efficiency of animals maintained on three dietary regimes (unit dry matter intake per unit gain)

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Treatments	Ration A	Ration B	Ration C
Number of animals	6	6 .	6
0-7 days	1.97	1.95	1.99
7-14	2.14	2.03	2.05
14-21	2.21	2.18	2.17
21-28	2.35	2.21	2.29
28-35.	2.54	2.43	.2.43
35-42	2.79	2.53	2,57
2-49	2.83	2.66	2.73
9-56	2.95	2.76	2.84
6-63	2.9 9	2.80	2.96
3-70	3.02	2.85	3.04
0-77	3.12	2.96	3.13
7-84	3.19	3.12	3.20

Table7. Average weekly protein efficiency of animals maintained on three dietary regimes (unit protein consumed per unit gain)

8. Summarised data on daily dry matter consumption, dry matter consumption per 100 kg body weight, average cumulative feed efficiency and average cumulative protein efficiency of goats maintained on three dietary regimes (Rations A, B and C)

<u> </u>			·
Treatments	Ration A containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202%	Ration B containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Ammonium chloride-1%	Ration C containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Horse gram extract 1 lit/animal/day
Number of animals	6	6	6
Average daily dry matter consumption (g)	928.57	917. 98	901.38
Dry matter consumption per 100 kg body weight	3.71	3.64	3.62
Average cumulative feed efficiency	15.02±0.141	14.58±0.109	14.69±0.150
Average cumulative protein efficiency	2.69±0.022	2.60±0.20	2.62±0.023

Table8 .

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Table 9	Average total erythrocyte count (TEC) (10° animals maintained on the experimental	/mm ³) of rations
	(Rations A, B and C)	

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Ration A	Ration B	Ration C
6	6 6	6
12,41±0,72	12.45±0.63	12.92±0.46
13.25±0.54	12.25±0.37	13.97±0.35
12.58±0.68	13.00±0.55	12.04±0.59
12.70±0.63	13.34±0.66	12.84±0.64
	6 12.41±0.72 13.25±0.54 12.58±0.68	6 6 12.41±0.72 12.45±0.63 13.25±0.54 12.25±0.37 12.58±0.68 13.00±0.55

Table 10. Average total leucocyte count (TLC) (10³/mm³) of animals maintained on the experimental rations (Rations A, B and C)

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	11.93±0.51	12.16±0.72	11.76±0.44
28 day	10.54±0.85	11.40±0.24	11.10±0.42
56 day	11.94±0.30	12.70±0.38	11.45±0.36
84 day	11.08±0.69	11.86±0.5 9	11.48±0.71

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	8.37 <u>±</u> 0.26	8.57±0.27	8.53±0.22
28 day	8.41±0.35	8.04±0.28	8,26±0.38
56 day	8.63±0.75	8.25±0.16	8.44±0.46
84 day	8.27±0.22	8.40±0.23	8.23±0.17

Table 11. Average haemoglobin concentration (g/dl) of animals maintained on the experimental rations (Rations A, B and C)

Table ₁₂. Average plasma protein concentration (g/dl) of animals maintained on the experimental rations (Rations A, B and C)

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	7.77±0.29	7.38±0.62	7.02±0.17
28 day	7.92±0.18	7.83±0.21	7.45±0.26
56 day '	7.49±0.26	7.26±0.63 '	7.57 <u>+</u> 0.43
84 day	8.09±0.36	7.43±0.48	7.79±0.46

Table 13. Summarised data on TEC, TLC, haemoglobin and plasma protein values of goats maintained on three experimental rations for a period of 84 days

Treatments	Ration A containing Calcium-1.194%	Ration B containing Calcium-1.194%	Ration C containing Calcium-1.194%
	Phosphorus-0.578 Magnesium-1.202%		Phosphorus-0.578% Magnesium-1.202% Horse gram extract 1 lit/animal/day
Number of animals	6	6	6 .
	TEC	(x10 ⁶ /mm ³)	
Initial (0 day)	12.41±0.72	12.45±0.63	12.92±0.46
Final (84 day)	12.70±0.63	13.34±0.66	12.84±0.64
Difference	0.29	0.89	0.08
	TLC	(x10 ³ /mm ³)	
Initial (0 day)		12.16±0.72	11.76±0.44
Final (84 day)	11.08±0.69	11.86±0.59	11.48±0.71
Difference	0.85	0.30	0.28
•	Наетод	lobin (g/dl)	
nitial (0 day)	8.37±0.26	8.57±0.27	8.53±0.22
inal (84 day)	8.27±0.22	8.40±0.23	8.23±0.17
ifference	0.10	0.17	0.30
	Plasma p	rotein (g/dl)	
nitial (0 day)	7.77±0.29	7.38±0.62	7.02±0.17
inal (84 day) ifference	8.09±0.36	7.43±0.48	7.97±0.46
llierence	0.32	0,05	0.95

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Table 1 4.	Average serum cal animals maintained (Rations A, B and	l'on the	centration experiment	(mg/dl) of cal rations
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Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	10.41±0.18	10.32±0.21	10.50±0.16
28 day	8.93±0.44	9.45±0.17	8.73±0.25
56 day .	8.27±0.12	8.79±0.28	8.31±0.36
84 day	7.08±0.23	8.63 <u>±</u> 0.20	8.32 <u>-</u> 0.11

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Table 15. Average serum inorganic phosphorus concentration (mg/dl) of animals maintained on the experimental rations (Rations A, B and C)

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	5.35±0.11	5.31 <u>+</u> 0.16	5.58±0.21
28 day	5.94±0.21	5.62±0.23	5.75 <u>±</u> 0.34
56 day	6.72 <u>+</u> 0.34	5.73±0.40	6.30±0.32
34 day	. 7.67 <u>±</u> 0.07	6.10 <u>+</u> 0.10	6.69±0.08

Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
0 day	2.60±0.12	2.53±0.20	2.67±0.17
28 day	4.78±0.28	3.41±0.26	3.99±0.28
56 day	5.65±0.21	4.16±0.32	4.71±0.36
84 day	6.08±0.15	4.70±0.14	5.50±0.12

Table 16. Average serum magnesium concentration (mg/dl) of animals maintained on the experimental rations (Rations A, B and C)

Table 17. Summarised data on serum calcium, serum inorganic phosphorus and serum magnesium values of goats maintained on three experimental rations for a period of 84 days

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Treatments	Ration A containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202%	Ration B containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Ammonium chloride-1%	Ration C containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Horse gram.extract 1 lit/animal/day
Number of . animals	6	6	б
	ŕ		
- · · ·	Serum cal	cium (mg/dl)	
Initial (0 day) Final (84 day)	10.41±0.18 7.08±0.23	10.32±0.21 8.63±0.20	10.50±0.16 8.32±0.11
Difference	3.33	1.69	2.18
	Serum inorganic	phosphorus (mg/dl)	• . •
Initial (0 day)	5.35±0.11	5.31±0.16	5.58±0.21
Final (84 day)	7.67±0.07	6.10±0.10	6.69±0.08
Difference	2.32	0.79	1.11
	Serum magne	esium (mg/dl)	· · ·
Initial (0 day)	2.60±0.12	2.53±0.20	2.67±0.17
Final (84 day)	6.08±0.15	4.70±0.14	5.50±0.12
Difference 🕐 🕚	3.48	2.17	2.83

Table 18. Data on calcium balance and per cent calcium retention of the animals in three groups maintained on conventional concentrate ration during initial (0 day) metabolism trial

Groups	I,	II	III
Number of animals	6	. 6	6
Calcium intake (g/day)	9.091	8.139	8.391
Calcium outgo			• •
Faecal (g/day)	3.352	2.925	3.147
Urinary (g/day)	0.104	0.109	0.108
(mg/dl)	7.542	7.334	, 7.956
Total (g/day)	3.456	3.034	3.247
Calcium balance	5.635	5.105	5.144
Per cent retention of calciúm	62.05	62.71	61.04

Table 19.. Data on calcium balance and per cent calcium retention of the animals in three groups maintained on three dietary regimes during second (28th day) metabolism trial

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Treatment	Ration A	Ration B	Ration C
Number of animals	· 6	6 ,	6
Calcium intake (g/day)	10.028	9.970	9.728
Calcium outgo			
Faecal (g/day)	3.760	3.551	3.594
Urinary (g/day)	0.134	0.119	0.124
(mg/dl)	8.201	6.324	5.701
Total (g/day)	3.894	3.670	3.718
Calcium balance	6.134	6.300	6.010
Per cent retention of calcium	61.17	63.19	61.78

Table 20.. Data on calcium balance and per cent calcium retention of the animals in three groups maintained on three dietary regimes during third (56th day) metabolism trial

Ration A	Ration B	Ration C
6	6	. 6
12.436	11.920	11.353
·		. 1
4.769	4.109	4.118
0.169	0.126	0.136
11.715	6.966	8.136
4.938	4.235	4.254
7.498	7.685	7.099
60.29	64.47	62.53
	6 12.436 4.769 0.169 11.715 4.938 7.498	6 6 12.436 11.920 4.769 4.109 0.169 0.126 11.715 6.966 4.938 4.235 7.498 7.685

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Table 21 . Data on calcium balance and per cent calcium retention of the animals in three groups maintained on three dietary regimes during final (84th day) metabolism trial

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Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6
Calcium intake (g/day)	13.245	13,349	12.129
Calcium outgo	· .		
Faecal (g/day)	5.115	4.304	4.472
Urinary (g/day)	0.191	0.128	0.151
(mg/dl)	12.862	5.587	7.968
Total (g/d a y)	5.305	4.432	4.622
Calcium balance	7.939	8.917	7.756
Per cent retention of calcium	59.85	66.55	62.65
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Table 22. Data on phosphorus balance and per cent phosphorus retention of the animals in three groups maintained on conventional concentrate ration during initial (0 day) metabolism trial

Groups	I	II	III
Number of animals	6	6	6
Phosphorus intake (g/day)	2.788	2.589	2.682
Phosphorus outgo	· .		
Faecal (g/day)	1.384	1.349	1.218
Urinary (g/day)	0.182	0.197	0.199
(mg/dl)	13.197	13.255	14.661
Total (g/day)	1.566	1.547	1.418
Phosphorus balance (g/day)	1.222	1.163	1.264
Per cent retention	46.64	45.28	47.48

Table 23. Data on phosphorus balance and per cent phosphorus retention of the animals in three groups maintained on three dietary regimes during second (28th day) metabolism trial

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Ration A	Ration B	Ration (
6	, 6	б.
4.954	4.985	4.775
	•	
2.572	2.538	2.461
0.290	0.286	0.231
18.237	12.011	. 10.621
2.870	2.764	2.692
2.084	2.221	2.083
42.07	44.56	43.63
	6 4.954 2.572 0.290 18.237 2.870 2.084	6 6 4.954 4.985 2.572 2.538 0.290 0.286 18.237 12.011 2.870 2.764 2.084 2.221

Table 24. Data on phosphorus balance and per cent phosphorus retention of the animals in three groups maintained on three dietary regimes during third (56th day) metabolism trial

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Treatment	Ration A	Ration B	Ration C
Number of animals	6	6	6
Phosphorus intake (g/day)	5.990	5.742	5.594
Phosphorus outgo	•.		··· .
Faecal (g/day)	3.502	3.460	2.996
Urinary (g/day)	0.438	0.239	0.258
(mg/dl)	30.362	13.214	15.434
Total (g/day)	3.925	3.221	3.254
Phosphorus balance	2.065	2.521	2.340
Per cent retention of phosphorus	34.47	43.90	41.83

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Data on phosphorus balance and per cent phosphorus of the animals in three groups maintained on three dietary regimes during final (84th day) metabolism Table 25. • • trial

Treatments	Ration A	Ration B	Ration C
Number of animals	· 6	6	6
Phosphorus intake (g/day)	6.593	6.626	6.243
Phosphorus outgo			
Faecal (g/day)	3.740	3.530	3,613
Urinary (g/day)	0.530	0.231	0.275
(mg/dl)	35.690	10.083	14,512
Total (g/day)	4.270	3.761	3.888
Phosphorus balance (g/day)	2.323	2.865	2.355
Per cent retention of phosphorus	35.24	43.24	37.72

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Table 26. Data on magnesium balance and per cent magnesium of the animals in three groups maintained on conventional concentrate ration during initial (0 day) metabolism trial

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Groups	I	II	III
Number of animals	6	6	6
Magnesium intake (g/day)	1.844	1.721	1.906
Magnesium outgo	. /	,	
Faecal (g/day)	1.003	0.977	1.033
Jrinary (g/day)	0.220	0.190	0.212
(mg/dl)	15.958	12.785	15.619
'otal (g/day)	1,224	1.166	1.246
agnesium balance g/day)	• 0.620	0.554	0.659
er cent retention f magnesium	33.20	31.86	34.00

Table 27 . Data on magnesium balance and per cent magnesium retention of the animals in three groups maintained on three dietary regimes during second (28th day) metabolism trial

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Treatment	Ration A	Ration B	Ration C
Number of animals	6	• 6 _.	6.
Magnesium intake (g/day)	10.028	9.987	9.563
Magnesium outgo			
Faecal (g/day)	5.621	5.782	5.221
Urinary (g/day)	0.785	0.653	0.761
(mg/dl)	48.042	34.703	34.988
Total (g/day)	6.406	6.435-	5.982
Magnesium balance	3.622	3.552	3.581
Per cent retention	36.12	35.57	37.45

Table 28 . Data on magnesium balance and per cent magnesium retention of the animals in three groups maintained on three dietary regimes during third (56th day) metabolism trial

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Treatment	Ration A	Ration B	Ration C
Number of animals	6	6	6
Magnesium intake (g/day)	12.316	11.921	11. 114
Magnesium outgo		- ·	
Faecal (g/day)	5.961	6.235	5.531
Urinary (g/day)	1.346	1.121	1.179
(mg/dl)	93.304	61.971	70.528
Total (g/day)	7.307	7.356	6.710
Magnesium balance	5.009	4.565	4.404
Per cent retention of magnesium	40.67	38.29	39.63

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Table 29. Data on magnesium balance and per cent magnesium retention of the animals in three groups maintained on three dietary regimes during final (84th day) metabolism trial

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Treatments	Ration A	Ration B	Ration C
Number of animals	6	6	6,
· · · · · · · · · · · · · · · · · · ·	· ·		
Magnesium intake (g/day)	13.512	13.565	13.022
Magnesium outgo			
Faecal (g/day)	6.101	6.503	5.973
Urinary (g/day)	1.617	1.580	1.590
(mg/dl)	108.889	68.966	. 83.905
Total (g/day)	7.718	8.084	7.563
Magnesium balance (g/day)	5.793	5.432	5.459
Per cent retention of magnesium	42.49	40.01	41.80

Table 30⁻. Data on nitrogen balance and per cent nitrogen retention of the animals in three groups maintained on three dietary regimes during second (28th day) metabolism trial

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Treatment	Ration A	Ration B	Ration C
Number of animals	6	6	6
Nitrogen intake (g/day)	28.379	27.375	26.146
Nitrogen outgo			
Faecal (g/day)	9.576	8.226	. 8.493
Urinary (g/day)	8.392	7.675	8.415
Total (g/day)	17.968	15.901	16.908
Nitrogen balance	10,415	10.531	8.942
Per cent retention of nitrogen	36.70	38.47	34.20

Table 31. Data on nitrogen balance and per cent nitrogen retention of the animals in three groups maintained on three dietary regimes during third (56th day) metabolism trial

Treatment	Ration A	Ration B	Ration C
Number of animals	6	6	6
Nitrogen intake (g/day)	32.497	35.898	32.874
Nitrogen outgo			
Faecal (g/day)	10.056	10.667	9.862
Urinary (g/day)	10.142	9.475	9.773
Total (g/day)	20.198	20.142	19.635
Nitrogen balance	12.502	14.782	12.430
Per cent retention of nitrogen	38.48	41.18	37.85

Table 32. Data on nitrogen balance and per cent nitrogen retention of the animals in three groups maintained on three dietary regimes during final (84th day) metabolism trial • . . .

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Ration A	Ration B	Ration C
6	6	6
39.544	41.064	37.457
	·	
11.610	11.997	11.665
11.445	10.953	10.907
23.060	22 .9 49	22.562
16.485	18,115	14.895
41.30	43.50	⁻ 39.76
	6 39.544 11.610 11.445 23.060 16.485	6 6 39.544 41.064 11.610 11.997 11.445 10.953 23.060 22.949 16.485 18.115

Table 33.	Summarised data on urine calcium,	phosphorus,
	magnesium and nitrogen of animals ma	aintained on
	three experimental rations	

Treatments	Ration A containing Calcium-1.194 Phosphorus-0. Magnesium-1.2	578% Phosphorus-0.578	
Number of animals	6	6	6
	c	alcium (g/day)	
Initial (0 day Final (84th da Difference		0.109 0.128 0.019	0.108 0.151 0.043
•.	Ph	osphorus (g/day)	
Initial (0 day Final (84th da Difference		0.197 0.231 0.034	0.199 0.275 0.076
- - -	Ма	gnesium (g/day)	·
Initial (0 day Final (84th da Difference) 0.220 y) 1.617 1.397	0.190 1.580 1.390	0.212 1.590 1.378
· · ·	N	itrogen (g/day)	
Initial (0 day Final (84th da Difference		6.160 10.953 4.793	6.618 10.907 4.289

 	Ration A containing Calcium-1.194 Phosphorus-0. Magnesium-1.2	578% Phosphorus-0.578%	Ration C containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Horse gram extrac 1 lit/animal/day
Number of animals	6	. 6	б
	С	alcium (g/day)	
	3.352	2,925	3.147
Initial (0 day) Final (84th day		4.304	4.472
Difference	1.763	1.379	1.325
	Ph	osphorus (g/day)	
Initial (0 day)	1.384	1.349	1,218
Final (84th day		3.530	3.613
Difference	2.356	2.181	2,395
	Ма	gnesium (g/day)	
Initial (0 day)	1.003	• 0.977	1.033
Final (84th day		6.503	5,973
Difference	5.098	5,526	4.940
	. N	itrogen (g/day)	
Initial (0 day)	6.904	6.319	6.854
Final (84th day		11.997	11.665
Difference	4.706	5.678	4.811

Table 34 . Summarised data on faecal calcium, phosphorus, magnesium and nitrogen of animals maintained on

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. ·	Ration A containing Calcium-1.194% Phosphorus-0.57 Magnesium-1.202	E	Ration C containing Calcium-1.194% Phosphorus-0.578% Magnesium-1.202% Horse gram extract 1 lit/animal/day
Number of animals	6	6	б.
	c	alcium (%)	
Initial (0 day) Final (84th day		62.71 66.55	61.04 62.65
Difference	2.2	3.84	1.61
	Pho	osphorus (%)	
Initial (0 day) Final (84th day	46.64) 35.24	45.28 43.24	47.48 37.72
Difference	11.40	2.56	9.76
-	Мас	gnesium (%)	
Initial (0 day)	32.20	31.86	34.00
Final (84th day) Difference	42.49 9.7	40.01 8.15	41.80 7.8
•	Ni	trogen (%)	
Initial (0 day)	32.60	34.35	31.43
Final (84th day) Difference	41.30 8.7	43.50 9.15	39.76 8.33

Table 35. Summarised data on per cent retention of animals maintained on three experimental rations



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