Seasonality in digestion and rumen metabolism in red deer (*Cervus elaphus*) fed on a forage diet

BY D. O. FREUDENBERGER[1]*, K. TOYAKAWA[1]†, T. N. BARRY[1]§, A. J. BALL[1]‡ AND J. M. SUTTIE[2]

[1]*Department of Animal Science, Massey University, Palmerston North, New Zealand,*
[2]*AgResearch Invermay, Mosgiel, New Zealand*

(*Received 27 January 1993 – Revised 5 July 1993 – Accepted 19 July 1993*)

Six adult castrated male red deer (*Cervus elaphus*), fitted with rumen cannulas, were offered chaffed lucerne hay *ad lib.* during winter and summer, with voluntary food intake (VFI) being respectively 59 and 89 g dry matter intake (DMI)/kg bodyweight 0·75 per d. The same animals were also offered the same feed during summer, with intake restricted to that of winter VFI. The apparent digestibility of gross energy (0·60) or fibre (0·41) and the total capacity (volume) of the rumen were unaffected by season or level of intake. Relative to winter *ad lib.* feeding, N retention, total rumen pool size (DM + water), rumen pool size as a proportion of capacity, and rumen total volatile fatty acid (VFA) pool size were increased during summer *ad lib.* feeding. Relative to winter *ad lib.* feeding, N retention, rumen ammonia irreversible loss rate (IRL), total rumen pool size, rumen pool size as a proportion of capacity, and rumen ammonia and total VFA pool sizes were also increased during summer restricted feeding. Rumen lignin fractional disappearance rate (FDPR) was lower in summer than in winter, and there was a non-significant trend for rumen fractional outflow rate (FOR) of liquid to follow the same trend. Molar proportions of acetate and propionate were unaffected by season, proportions of *n*-butyrate were slightly higher in summer, and proportions of iso-butyrate and iso-valerate were higher for summer restricted than for winter *ad lib.* feeding. When intakes were equalized there were no seasonal changes in rate of rumen water outflow, net rumen water balance or intestinal water absorption. It is concluded that there is a seasonal change in rumen physiology in red deer during summer causing increased total rumen pool size (DM + water), an increase in rumen ammonia production and pool size, and an increase in rumen total VFA pool size which are all independent of the increase in VFI. The increased total rumen pool size in the summer restricted group may indicate an increased mean retention time (MRT) of digesta in the rumen. MRT for particulate matter was calculated to be 29·2 and 34·8 h during winter and summer respectively. This, together with increased rumen ammonia production, may function to maintain rumen fibre digestion when VFI normally increases during summer. The increased rumen VFA pool size may indicate increased VFA production during summer, in the same way as ammonia IRL was increased.

Red deer: Digestion: Rumen pool size: Ammonia production: Rumen outflow

Deer from temperate regions show a marked seasonality in voluntary feed intake (VFI) and body growth, with lower values in winter and higher values in summer (Milne *et al.* 1978; Barry *et al.* 1991). The summer increase in VFI is also associated with a marked increase in rumen digesta load, but there is hypertrophy of the rumen tissues (Sibbald & Milne 1993). The objectives of the present investigation were to establish whether the red deer

* Present address: CSIRO Division of Wildlife and Ecology, PO Box 84, Lyneham, ACT, Australia.
† Present address: Laboratory of Animal Science, Faculty of Agriculture, Hirosaki University, Hirosaki, Japan.
‡ Present address: Department of Animal Science, University of New England, Armidale, Australia.
§ For reprints.

(*Cervus elaphus*) also shows seasonal changes in digestive function and to establish whether any seasonal changes were independent of seasonal changes in VFI. Criteria investigated were apparent digestibility, rumen pool size and fractional outflow rate (FOR), N excretion and retention, rumen volatile fatty acid (VFA) concentration, pool size and molar proportions and rumen NH_3 production, here defined as the irreversible loss rate (IRL).

MATERIALS AND METHODS

Experimental design

One experiment with three treatments was conducted, each with the same format and using the same animals and feed from the same batch. The first treatment was conducted at *ad lib.* intake during winter, the second at *ad lib.* intake in summer and the third during summer with intake restricted to the same level as recorded for winter *ad lib.* intake (summer restricted). Each experimental period comprised a 2 week adjustment period to the diet followed by a 7 d collection period to measure the apparent digestibility of the feed. An indigestible marker was then infused into the rumen for a further 5 d and ^{15}N-labelled NH_4Cl was added for the last 2 d during treatments 1 and 3, to determine rumen FOR and rumen NH_3 IRL respectively. Rumen fluid samples were removed on 3 d during the infusion to determine the concentrations of VFA and NH_3, and to check plateau attainment for ^{15}N enrichment of NH_3. Rumen contents were removed by baling at the end of the infusion to determine pool sizes and ^{15}N enrichment of NH_3.

Animals, housing and diets

Six castrated hand-reared male red deer aged 3–5 years were used. All were fistulated in the rumen and fitted with a permanent rubber cannula (83 mm i.d.), were well accustomed to the experimental conditions, and were kept in metabolic crates similar to those described by Milne *et al.* (1978). There was no leakage from the rumen cannulas during any of the three treatment periods. The crates were housed in a well ventilated building with artificial light set at 9 h light–15 h dark during the winter period and 15 h light–9 h dark during the two summer periods. Water was provided *ad lib.* and a multi-mineral salt block (Dominion Salt, Blenheim, NZ) was in each feed bin at all times. Good quality lucerne (*Medicago sativa*) hay, all from the same batch, was chopped into 10–30 mm lengths and given in each period at hourly intervals from overhead feeders.

Treatments

During the first period (winter *ad lib.*, June 1990) feed was offered *ad lib.* (1·2 times the previous day's consumption) for 2 weeks. During the subsequent weeks feed was offered at 1·035 of voluntary feed intake (VFI) in order to minimize diet selectivity. During the second period (summer *ad lib.*, Feb 1991) feed was offered exactly as described for winter *ad lib.* feeding. During the final period (summer restricted; March 1991) feed was offered at 1·035 of the winter VFI on a g dry matter (DM)/kg bodyweight $(W)^{0·75}$ per d basis, as liveweight had increased slightly from winter to summer.

Marker infusion

The inert fluid marker Cr-EDTA was prepared by the method of Binnerts *et al.* (1968) and adjusted to a pH of 6·5–8·0. The Cr-EDTA was made up to 20 l with a final Cr concentration of 2 mg/g of solution. Following a priming dose of 40 g into the rumen, the marker solution was continuously infused into the rumen for 5 d at a rate of 23–25 g/h. The exact infusion rate was determined for each animal. The infusion was administered by a

peristaltic pump (PLG-multipurpose pump; Desaga, Heidelberg, Germany). ^{15}N-labelled NH_4Cl solution (99·0 atoms % excess, 0·5 mg/g of infusate; Amersham, UK) was added to the Cr-EDTA solution during the last 45 h of infusion.

Sample collection

Feed, feed refusals and faeces were weighed daily during the 7 d collection period and a sub-sample (20% of each material) was taken and stored at −20°. Another daily sub-sample of each material was taken for duplicate determination of DM in a forced draught oven (100°). Urine was weighed daily from buckets containing sufficient H_2SO_4 (250 ml/l) to maintain pH below 3·5. After the collection periods all frozen subsamples were bulked within animals, mixed thoroughly and re-sampled, then freeze dried, ground through a 1 mm mesh sieve (Wiley Mill, USA) and used for analysis.

Rumen fluid samples for VFA, NH_3-N concentration and Cr determinations were taken twice daily (10·00 and 15·00 hours) on 3 d during the infusion period, and also at rumen emptying. During the infusion period the samples were taken from a tube that passed through the bung of the rubber cannula and was attached to a perforated brass cylinder covered with nylon mesh (80 micron aperture; Swiss Screens, Sydney, Australia). The length of the sampling tube was adjusted so that the sampling apparatus hung within the middle of the rumen. The first 20 ml of rumen fluid was discarded at each sampling time. Rumen fluid was also obtained when the rumen was emptied by squeezing mixed digesta through a nylon mesh (80 micron aperture). Fluid for NH_3-N and VFA analysis was acidified, deproteinized and centrifuged as described by Domingue *et al.* (1991*a*).

The rumens were emptied while the deer were slightly sedated with xylazine (Rompun, Bayer AG, Germany). The rumen contents were weighed, mixed thoroughly and subsampled before returning the warmed digesta back to the rumen. The procedure took 15–20 min per animal and the animals remained standing throughout.

Rumen fluid was sampled for ^{15}N enrichment at 23, 35 and 45 h after commencing the infusion. This final sample (45 h) was taken when the rumen was emptied and its contents mixed. These rumen fluid samples were immediately acidified with 0·1 ml concentrated H_2SO_4, centrifuged, and the supernatant stored at −20°.

Rumen capacity

Rumen capacity (i.e. volume) was determined by inflating, with warm water, a balloon placed inside the emptied rumen as described by Domingue *et al.* (1992). This measurement was made only at the end of the winter *ad lib.* and summer restricted periods.

Analytical

Samples of feed, feed refused, faeces and rumen digesta were analysed for cell wall constituents by the detergent method (not including the amylase) of Robertson & Van Soest (1980), total N by the Kjeldahl method, gross energy by adiabatic bomb calorimetry (Gallenkamp Autobomb; Loughborough, Leics.), and organic matter by ashing overnight at 550°. Total N of urine was also determined by the Kjeldahl method. Cr concentration was determined by atomic absorption spectrometry. NH_3-N and VFA were determined as described by Domingue *et al.* (1991*a*).

^{15}N-NH_3 was isolated from rumen fluid samples by the use of an ion-exchange column as modified from Read *et al.* (1982). Acidified rumen fluid was centrifuged at 9000 rev./min, and a 5 ml portion was adjusted to pH 5·5–7·5 with 1 M-NaOH; Na-K cation exchange resin (Dowex 50, 100–200 mesh) suspension (2 ml) was then added to the rumen fluid preparation and mixed for 15 min. The supernatant was discarded and the resin was washed three times with deionised water. NH_3 was eluted off the resin with 2·5 M-$KHSO_4$

and this supernatant was evaporated to dryness. ^{15}N enrichment of the resulting ammonium bisulphate was determined by mass spectrometry (±0·0002 atoms %; Waikato Stable Isotope Unit; Hamilton, New Zealand).

Calculations

Fractional outflow rate (FOR) and fractional disappearance rates (FDPR) from the rumen were calculated as shown below. The method used to determine FOR is the continuous infusion and total sampling procedure (Faichney, 1975).

$$\text{FOR (\% per h)} = \frac{\text{Marker infusion rate (mg/h)} \times 100}{\text{Rumen pool size (mg)}}, \quad (1)$$

$$\text{FDPR (\% per h)} = \frac{\text{Intake (g/h)} \times 100}{\text{Rumen pool size (g)}}. \quad (2)$$

Cr-EDTA was used to calculate rumen liquid FOR and lignin to calculate particulate FOR; in the latter case, faeces lignin excretion was substituted for infusion rate in equation (1), on the assumption that there was minimal post-ruminal degradation. Rumen pool sizes were determined when the rumens were emptied.

The IRL of NH_3-N was expressed as the rate (mass/unit time) at which NH_3-N leaves the rumen pool and does not return during the experimental period (Nolan & Leng, 1972):

$$\text{IRL (mg N/d)} = \frac{\text{infusion rate of } ^{15}N \text{ (mg/d)}}{\text{enrichment at plateau of rumen } NH_3\text{-N with } ^{15}N}. \quad (3)$$

Plateau enrichment used in equation (3) was determined from samples taken when the rumen was emptied, after 45 h of ^{15}N infusion. Rumen NH_3-N outflow was calculated by multiplying rumen NH_3-N pool size by the FOR of Cr-EDTA. The outflow of water from the rumen was calculated as rumen liquid pool (g) × FOR of Cr-EDTA (d). Rumen water balance was calculated as the difference between rumen water outflow and total water intake (g/d); it represents the combined total of salivary secretion and net inflow of water across the rumen wall. Apparent absorption of water from the intestines was calculated as the difference between rumen water outflow and faeces water output.

Statistical analysis

Mean values and the standard errors of differences of the means (SED) are presented. The values for NH_3-N and VFA are the means of seven observations/animal, since there were no significant differences ($P > 0·1$) between the day, time of day or type of sampling (rumen emptying or by sampling apparatus). Univariate analysis of variance of repeated measures (Gill, 1986; Genstat Statistical Software, 1988) was used to remove between-animal variation before analysing for seasonal differences. Means were compared by the method of least significant difference (Snedecor & Cochran, 1980).

RESULTS

Chemical composition of the lucerne chaff fed showed only minor variation between the three feeding periods (Table 1). Mean liveweight of the deer during summer *ad lib.* feeding (109·7 kg) was significantly higher ($P < 0·001$) than during winter *ad lib.* feeding (96·1 kg). Mean liveweight during summer restricted feeding (104·5 kg) was also significantly higher than during winter *ad lib.* feeding ($P < 0·01$). VFI of all the constituents measured (Table 2) were much higher for summer *ad lib.* than for winter *ad lib.* feeding ($P < 0·001$), whilst

Table 1. *Composition (g/kg dry matter (DM)) of chopped lucerne* (Medicago sativa) *hay fed to rumen fistulated red deer* (Cervus elaphus) *stags during winter and summer* ad lib. *and summer restricted periods**

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted
Organic matter	907	891	877
Neutral detergent fibre	407	380	382
Acid detergent fibre	279	256	252
Lignin	58	55	54
Total N	32·5	32·8	35·6
Gross energy (kJ/g DM)	19·0	18·2	18·0

* For details of treatments, see p. 490.

Table 2. *Voluntary intakes and digestible intakes of dry matter (DM), organic matter (OM) and fibre (neutral detergent residue) with their apparent digestibilities, of chopped lucerne* (Medicago sativa) *hay fed to rumen fistulated red deer* (Cervus elaphus)

(Mean values and standard error of the difference between means for six animals)

Feeding group*...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
Voluntary intake				
DM (g/kg $W^{0·75}$ per d)	58·7	88·6	61·3	1·90
(g/d)	1773	2988	1989	96·3
OM (g/kg $W^{0·75}$ per d)	53·2	76·5	56·1	2·55
(g/d)	1607	2576	1825	89·5
Fibre (g/kg $W^{0·75}$ per d)	22·9	30·7	24·3	1·07
Apparent digestibility				
DM	0·618	0·606	0·577	0·0124
OM	0·640	0·632	0·642	0·0198
Fibre	0·436	0·394	0·421	0·0337
Gross energy	0·609	0·600	0·614	0·0207

W, bodyweight.
* For details of procedures, see pp. 490–491.

the objective of maintaining intake of the summer restricted group to the same level as the winter *ad lib.* group on a per kg $W^{0·75}$ basis was attained. Apparent digestibility of DM was lower for summer restricted than for either winter or summer *ad lib.* feeding ($P < 0·05$). None of the treatments significantly affected apparent digestibility of organic matter (OM), energy and fibre.

None of the treatments affected total rumen capacity (Table 3). Relative to winter *ad lib.* feeding, summer *ad lib.* feeding resulted in greater rumen pool size per kg $W^{0·75}$ of both DM and liquid ($P < 0·01$) and markedly increased total (DM and liquid) and DM pools as a proportion of total rumen capacity ($P < 0·01$). Relative to winter *ad lib.* feeding, summer restricted feeding also increased total rumen pool size per kg $W^{0·75}$ ($P < 0·01$), total rumen pool size as a proportion of rumen capacity ($P < 0·05$) and DM pool as a proportion of capacity ($P < 0·05$). Differences in rumen pool sizes between summer restricted and winter *ad lib.* feeding were smaller in magnitude than the differences between summer *ad lib.* and winter *ad lib.* feeding. When values were expressed as kg/kg dry matter intake (DMI), summer restricted feeding increased total and liquid rumen pool sizes ($P < 0·01$) relative to winter *ad lib.* feeding, with summer *ad lib.* feeding being intermediate.

Table 3. *Rumen capacity, pool sizes and fractional outflow rates of Cr-EDTA and lignin from the rumen of red der* (Cervus elaphus) *stags fed on chopped lucerne* (Medicago sativa) *hay*†

(Mean values and standard error of the difference between means for six animals)

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
Rumen capacity (g H_2O/kg $W^{0.75}$)	487	509	447	31·3
Rumen pool size (g/kg $W^{0.75}$)				
Dry matter (DM)	35·9	57·1	39·2	2·90
Liquid	207	336	258	11·9
Total*	243	392	297	13·2
Rumen pool size (kg/kg DMI)				
DM	0·624	0·646	0·638	0·0478
Liquid	3·58	3·80	4·25	0·171
Total*	4·20	4·45	4·89	0·195
Rumen pool:rumen capacity				
DM pool:capacity	0·0745	0·1136	0·0873	0·0059
Total pool:capacity	0·496	0·776	0·670	0·039
Fractional disappearance rate (% per h)				
Lignin	3·43	2·90	2·84	0·349
Fractional outflow rate (% per h)				
Cr-EDTA	16·3	ND	12·4	2·52
Lignin	3·15	2·67	2·78	1·000
Cr-EDTA/lignin	5·37	ND	4·50	0·440
DM of rumen digesta (%)	14·7	14·6	13·2	0·79

W, Bodyweight; ND, not determined; DMI, dry matter intake.
* DM + liquid pools.
† For details of procedures, see pp. 490–492.

Table 4. *Nitrogen (N) excretion and retention in rumen fistulated red deer* (Cervus elaphus) *fed on chopped lucerne* (Medicago sativa) *hay**

(Mean values and standard error of the difference between means for six animals)

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
N fluxes (g N/kg $W^{0.75}$ per d)				
Intake	1·98	3·03	2·19	0·068
Faecal excretion	0·50	0·86	0·64	0·029
Urinary excretion	1·18	1·53	1·14	0·061
N balance				
g N/kg $W^{0.75}$ per d	0·30	0·63	0·41	0·047
g N/100 g DMI	0·51	0·71	0·67	0·075
g N/100 g N intake	15·1	20·7	18·7	2·18
Apparent N digestibility	0·749	0·714	0·708	0·1000
Urine excretion (proportion N intake)	0·598	0·507	0·521	0·0207

W, Body weight; DMI, dry matter intake.
* For details of procedures, see pp. 490–492.

Rumen lignin FDPR (Table 3) tended to be similar for both summer groups and lower than for the winter *ad lib.* group, with the difference between summer restricted and winter *ad lib.* groups attaining significance at $P < 0·1$. When data for both summer groups were pooled, the combined summer value for rumen lignin fractional disappearance rate differed from the winter *ad lib.* group at $P = 0·078$.

Table 5. *Concentration of ammonia, pool size and irreversible loss of* ^{15}N *in the rumen of red deer* (Cervus elaphus) *fed on chopped lucerne* (Medicago sativa) *hay**

(Mean values and standard error of the difference between means for six animals)

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
Ammonia				
Concentration (mg NH_3-N/l)	193	191	218	19·7
Pool size (g NH_3-N)	1·22	ND	1·83	0·094
Pool size (mg/g N intake)	20·4	ND	23·9	1·07
Outflow (mg N/g N intake)	77·3	ND	75·4	12·20
Irreversible loss rate (mg N/g N intake)	522	ND	649	29·1

ND, not determined

* For details of procedures, see pp. 490–492.

Rumen FOR of both Cr-EDTA and lignin also tended to be lower for summer restricted than for winter *ad lib.* feeding, but the differences did not attain significance ($P > 0{\cdot}10$). Although the combined summer value for rumen lignin FOR was lower than for the winter *ad lib.* value (2·73 *v.* 3·15%/h), the difference was not significant ($P = 0{\cdot}15$) due to the variation encountered.

Apparent N digestibility ($P < 0{\cdot}05$) and urinary N excretion (expressed as a proportion of N intake; $P < 0{\cdot}01$) were lower for both summer treatments than for the winter *ad lib.* treatment (Table 4). N retention of deer fed *ad lib.* during summer was higher than that of deer fed *ad lib.* during winter, regardless of whether this was expressed per kg $W^{0{\cdot}75}$ ($P < 0{\cdot}01$) or per 100 g DMI or N intake ($P < 0{\cdot}05$). N retention of deer on summer restricted feeding was intermediate, and significantly different from winter *ad lib.*-fed deer when expressed per kg $W^{0{\cdot}75}$ ($P < 0{\cdot}05$) and per 100 g DMI ($P < 0{\cdot}10$). Rumen NH_3 concentration and rumen NH_3 outflow were not affected by season of feeding (Table 5). Rumen NH_3 ^{15}N-enrichment attained plateau values before 45 h of infusion in both the summer and winter periods, and rumen NH_3 pool size ($P < 0{\cdot}05$) and IRL ($P < 0{\cdot}01$) were both higher during summer restricted than during winter *ad lib.* feeding (+50 and 24% respectively).

Drinking water and total water intakes were higher for both summer *ad lib.* ($P < 0{\cdot}001$) and summer restricted ($P < 0{\cdot}01$) than for winter *ad lib.* feeding (Table 6). However, there were no differences between winter *ad lib.* and summer restricted in net rumen water balance, rumen water outflow, intestinal water absorption, and faecal water efflux. Lower urinary efflux during summer ($P < 0{\cdot}01$) probably reflected greater body evaporative water losses at this time.

Relative to winter *ad lib.*, summer *ad lib.* feeding elevated rumen total VFA concentration ($P < 0{\cdot}05$) and pool size (per kg $W^{0{\cdot}75}$, $P < 0{\cdot}001$; per 100 g DMI, $P < 0{\cdot}05$), and slightly increased the molar proportions of *n*-butyrate ($P < 0{\cdot}01$; Table 7). These effects were still evident when summer VFI was reduced to the same level as winter VFI, but of a lower magnitude, with VFA concentration ($P < 0{\cdot}1$) and pool size (per kg $W^{0{\cdot}75}$ and per 100 g DMI, $P < 0{\cdot}05$) of summer restricted being higher than for winter *ad lib.*, and proportions of *n*-butyrate being higher for summer restricted than for winter *ad lib.* ($P < 0{\cdot}01$). Molar proportions of iso-butyrate and iso-valerate were greater for summer restricted than for winter *ad lib.* feeding ($P < 0{\cdot}05$).

Table 6. *Water fluxes and net rumen water balance in red deer* (Cervus elaphus) *fed on chopped lucerne* (Medicago sativa) *hay**

(Mean values and standard error of the difference between means for six animals)

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
Water fluxes (g/kg $W^{0.75}$ per d)				
Drinking influx	173	271	232	14·4
Total influx†	175	275	233	14·5
Net rumen water balance‡	635	ND	529	127·4
Rumen outflow§	807	ND	761	126·2
Intestinal absorption‖	765	ND	711	125·4
Faecal efflux	42	68	50	4·4
Urinary efflux	103	94	97	1·8
Faecal dry matter	0·345	0·328	0·356	0·0207

W, Bodyweight; ND, not determined.
* For details of procedures, see pp. 490–492.
† Drinking water + water in feed.
‡ Rumen outflow–total influx = salivary secretions + net flux of water across the rumen wall.
§ Rumen liquid pool × FOR (/d) of Cr-EDTA.
‖ Apparent absorption; rumen water outflow–faecal water efflux.

Table 7. *Concentration and molar proportions of volatile fatty acids (VFA) in the rumen of red deer* (Cervus elaphus) *fed on chopped lucerne* (Medicago sativa) *hay**

(Mean values and standard error of the difference between means for six animals)

Feeding group...	Winter *ad lib.*	Summer *ad lib.*	Summer restricted	SED
Total VFA				
Concentration (mmol/l)	81·9	96·5	91·0	5·47
Pool size (mmol/kg $W^{0.75}$)	16·9	32·4	23·6	2·23
(mmol/100 g DMI)	29·2	36·7	38·5	2·95
Molar proportion (mol%)				
Acetate	71·4	69·7	70·2	0·45
Propionate	18·4	18·4	17·7	0·31
N-Butyrate	6·4	7·7	7·5	0·31
Iso-butyrate	1·37	1·59	1·70	0·170
n-Valerate	1·01	1·07	1·11	0·066
Iso-valerate	1·38	1·62	1·79	0·181
Acetate:propionate ratio	3·89	3·81	3·98	0·079

W, Bodyweight; DMI, dry matter intake.
* For details of procedures, see pp. 490–491.

DISCUSSION

All temperate deer species show seasonality (Barry *et al.* 1991), which is manifest as annual cycles of VFI, body growth, plasma hormone concentrations, reproductive activity and coat type. The VFI cycle reaches a peak in summer and a trough in winter. Relative to the large amount of information on these aspects, there have been few studies on seasonality in digestive function in deer. Mean retention time (MRT) in the whole alimentary tract is shorter for red deer (*Cervus elaphus*) and Japanese sika deer (*Cervus nippon*) than for domesticated sheep, and this is associated with slightly lower apparent digestibility of organic matter and fibre in deer (Milne *et al.* 1978; Katoh *et al.* 1991). Domingue *et al.* (1991*b*) showed that rumen liquid FOR (i.e. 1/MRT) was faster for red deer (16% per h)

than for sheep and goats (10% per h), and that this occurred in both summer and winter. The present study has shown an increase in total rumen pool size and in the rumen pool sizes of liquid, NH_3 and VFA in summer compared with winter which are independent of VFI. The most logical explanation for the increased total and liquid pool sizes in summer restricted compared with winter *ad lib.*-fed deer is reduced rumen FOR of the liquid and particulate phases in summer. Although these did decline in this direction for both Cr-EDTA and lignin in the present study, the differences did not attain significance. However, rumen lignin fractional disappearance rate (FDPR) was shown to be lower in summer than in winter, and is related to fractional outflow rate (FOR) and fractional degradation rate (FDR) by equation 4:

$$\text{FDPR} = \text{FOR} + \text{FDR}. \quad (4)$$

In the present study there was minimum degradation of lignin (Table 3), with FDPR being of similar magnitude to FOR, but less variable to measure, thus suggesting that rumen outflow rate of particulate matter must have been slower in summer than in winter.

In the present study, apparent rumen MRT of particulate matter (1/lignin FDPR) can be calculated as 29·2 and 34·8 h for winter and summer. Similar values of 28·8 h for winter and 36·1 h for summer can be calculated from Domingue *et al.* (1991*b*) for true particulate MRT (1/lignin FOR), for red deer fed on a similar diet to that used in the present study. As total rumen volume (i.e. capacity) did not change between seasons, this may be a digestive adaptation that deer have evolved to increase rumen MRT (and hence time for microbial attack) under conditions where VFI increases in summer, thus ensuring that apparent digestibility does not decline with the summer increase in VFI.

This is the first study to show a seasonal increase in the rate of rumen NH_3 production, and we have shown this to be independent of changes in VFI. This may be one of the components responsible for maintaining fibre digestion as VFI increases during summer, especially if the diet is high in lignin and low in N, such as the *Agrostis-Festuca* and heather diets fed by Milne *et al.* (1978). The mechanism of the increased rumen NH_3 IRL presumably involves increased recycling of N into the rumen during summer, as dietary N intakes were similar in the winter and summer restricted groups. Iso-butyrate and iso-valerate are formed from deamination of valine and leucine in the rumen (Van Soest, 1982). The increased concentration of these compounds in the rumen fluid of summer restricted deer also suggests that protein degradation rate may be faster in summer than in winter, also contributing to the greater NH_3 IRL.

It seems that deer show a seasonal increase in rumen total VFA pool size during summer that is independent of changes in VFI. As rumen liquid volume was greater in summer than in winter, contributing causes may be longer residence time of material in the rumen during summer, giving more time for fermentation activity, and perhaps a greater rate of fermentation. A combination of the two could potentially result in a greater rate of VFA production during summer, as observed for rumen NH_3 production (i.e. IRL), and this should be studied in future experiments.

Increased N retention occurred from winter to summer during *ad lib.* feeding, as also found by Domingue *et al.* (1991*a*), but in the present study a component of this was independent of changes in VFI. It may be due to increases during late spring and summer in plasma concentrations of growth hormone (GH) and IGF-1 (Suttie *et al.* 1989) and prolactin (Brinklow & Forbes, 1982) and insulin secretory variables as evidenced by insulin clearance after the glucose tolerance test (IVGTT) (McMahon *et al.* 1992). The hormonal milieu in spring-summer of high GH, IGF-1 and prolactin and a large, rapid insulin response to the IVGTT followed by a rapid clearance of insulin and glucose is conducive, generally, to the effective deposition of protein. Whether this suitable hormonal milieu is a consequence of, or the cause of, the changes in VFI and rumen dynamics is not known.

The present study raises the possibility that many of the changes seen in deer, as seasonal growth rates vary, are endogenous and not simply a consequence of VFI. This may mean that seasonal cycles of growth and VFI, although functionally and intimately related, are not strictly identical.

Domingue *et al.* (1991*a*) observed a seasonal cycle in water metabolism in red deer, with increased rumen outflow, net rumen water balance (net inflow from saliva and diffusion) and intestinal absorption occurring in summer compared with winter. However, there were no differences in rumen water outflow and intestinal water absorption between winter and summer restricted feeding in the present study, showing these effects to be due only to increased VFI during summer. Net rumen water balance in fact decreased slightly in the summer restricted group, to counteract the increase from drinking at this time.

It is concluded that red deer show a seasonal cycle of rumen digestive function, with increased total rumen pool size, increased pool sizes of liquid, VFA and NH_3 and increased rumen NH_3 production in summer compared with winter, that are independent of changes in VFI. These indicate increased rumen retention time in summer and may function to maintain rumen fibre digestion rates when VFI normally increases during summer. The increased VFA pool size may indicate increased rumen VFA production during summer in the same way that rumen NH_3 IRL is increased.

Mr G. S. Purchas, Dr A. M. Ataja and Miss P. S. Parker are thanked for assistance with animal feeding and animal measurements, and Miss R. A. Watson, Mr D. A. Hamilton and Mrs Ai Hua are thanked for laboratory analyses. Mr W. Müller, CSIRO Biometrics Unit, is thanked for statistical advice.

REFERENCES

Barry, T. N., Suttie, J. M., Milne, J. A. & Kay, R. N. B. (1991). Control of food intake in domesticated deer. In *Physiological Aspects of Digestion and Metabolism in Ruminants*, pp. 385–402 [T. Tsuda, Y. Sasaki and R. Kawashima, editors). San Diego: Academic Press.

Binnerts, W. T., van't Klooster, A. T. & Frens, A. M. (1968). Soluble chromium indicator measured by atomic absorption in digestion experiments. *Veterinary Record* **82**, 470.

Brinklow, B. R. & Forbes, J. M. (1982). Prolactin infusion causes increased nitrogen retention in lambs in continuous darkness. *Proceedings of the Nutrition Society* **42**, 38A.

Domingue, B. M. F., Dellow, D. W., Wilson, P. R. & Barry, T. N. (1991*a*). Nitrogen metabolism, rumen fermentation, and water absorption in red deer, goats and sheep. *New Zealand Journal of Agricultural Research* **34**, 391–400.

Domingue, B. M. F., Dellow, D. W., Wilson, P. R. & Barry, T. N. (1991*b*). Comparative digestion in deer, goats and sheep. *New Zealand Journal of Agricultural Research* **34**, 45–53.

Domingue, B. M. F., Wilson, P. R., Dellow, D. W. & Barry, T. N. (1992). Effects of subcutaneous melatonin implants during long daylength on voluntary feed intake, rumen capacity and heart rate of red deer (*Cervus elaphus*) fed on a forage diet. *British Journal of Nutrition* **68**, 77–88.

Faichney, G. J. (1975). The use of markers to partition digestion within the gastro-intestinal tract of ruminants. In *Digestion and Metabolism of the Ruminant*, pp. 277–291 [I. W. McDonald and A. C. I. Warner, editors]. Armidale: University of New England Publishing Unit.

Genstat Statistical Software (1988). Genstat 5. Statistics Dept, Rothamsted Experimental Station, Hertfordshire, UK.

Gill, J. L. (1986). Repeated measurement: Sensitive tests for experiments with few animals. *Journal of Animal Science* **63**, 943–954.

Katoh, K., Kajita, Y., Odashima, M., Ohta, M. & Sasaki, Y. (1991). Passage and digestibility of lucerne (*Medicago sativa*) hay in Japanese sika deer (*Cervus nippon*) and sheep under restricted feeding. *British Journal of Nutrition* **66**, 399–405.

McMahon, C. D., Corson, I. D., Johnstone, P. D., Stuart, S. K., Veenvliet, B. A. & Suttie, J. M. (1992). Seasonal insulin response to an intravenous challenge in red deer stags. A possible role in the seasonal growth pattern? *Proceedings of the Australian Endocrinology Society* **35**, 164.

Milne, J. A., MacRae, J. C., Spence, A. M. & Wilson, S. (1978). A comparison of the voluntary intake and digestion of a range of forages at difference times of the year by the sheep and the red deer (*Cervus elaphus*). *British Journal of Nutrition* **40**, 347–357.

Nolan, J. V. & Leng, R. A. (1972). Dynamic aspects of ammonia and urea metabolism in sheep. *British Journal of Nutrition* **27**, 177–194.
Read, W. W. C., Harrison, R. A. & Halliday, D. (1982). A resin-based method for the preparation of molecular nitrogen for ^{15}N analysis from urinary and plasma components. *Analytical Biochemistry* **123**, 249–254.
Robertson, J. B. & Van Soest, P. J. (1980). The detergent system of analysis and its application to human foods. In *The Analysis of Dietary Fibre in Foods*, pp. 123–158 [W. P. T. James and O. Theander, editors]. New York: Marcel Dekker.
Sibbald, A. M. & Milne, J. A. (1993). Physical characteristics of the alimentary tract in relation to seasonal changes in voluntary food intake by the red deer. *Journal of Agricultural Science, Cambridge* **120**, 99–102.
Snedecor, G. W. & Cochran, W. G. (1980). *Statistical Methods*, 7th ed. Ames, Iowa: Iowa State University Press.
Suttie, J. M., Fennessy, P. F., Corson, I. D., Laas, F. J., Crosbie, S. F., Butler, J. H. & Gluckman, P. D. (1989). Pulsatile growth hormone, insulin-like growth factors and antler development in red deer stags. *Journal of Endocrinology* **121**, 351–360.
Van Soest, P. J. (1982). *Nutritional Ecology of the Ruminant*. Corvallis, Oregon, USA: O & B Books.

Printed in Great Britain