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Forage fibre digestion, rates of feed passage and gut fill in juvenile and adult red kangaroos *Macropus rufus* Desmarest: why body size matters

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Summary

Using red kangaroos *Macropus rufus* Desmarest, a large (>20 kg) marsupial herbivore, we compared the digestive capabilities of juveniles with those of mature, non-lactating females on high-quality forage (chopped lucerne *Medicago sativa* hay) of 43±1% neutral-detergent fibre (NDF) and poorer quality, high-fibre forage (chopped oaten *Avena sativa* hay) of 64±1% NDF. On chopped lucerne apparent dry matter (DM) digestibilities by young-at-foot (YAF) red kangaroos (an age that would normally be taking some milk from their mother), weaned juveniles and mature females were similar (55–59%). On chopped oaten hay apparent DM digestibility was lower in the YAF (35.9±2.3%) followed by weaned (43.4±2.8%) and mature females (44.6±1%). The digestion of NDF and its components (mainly cellulose and hemicellulose) was lowest among the YAF followed by weaned and then mature females. The YAF and weaned kangaroos could not sustain growth on the poor-quality diet, and appeared to be at or near maximal gut fill on both forages; the values being 114–122 g DM for YAF and 151–159 g DM for weaned kangaroos. Mean retention times (MRT) of particle and solute markers were significantly longer for the YAF and weaned kangaroos on oaten hay than on lucerne hay, and DM intake (g d^{-1}) was ~50% lower on the oaten hay. In contrast, solute and particle MRTs in the mature females were not significantly affected by diet; they maintained DM intakes by increasing DM gut fill from 264±24 g on chopped lucerne to 427±26 g DM on chopped oaten hay. Clearly, the mature female kangaroos did not maximise gut fill on the high-quality forage, presumably as a consequence of their proportionally lower energy requirements compared with still-growing juveniles. Overall, we have provided the first mechanistic link between the physiological constraints faced by juvenile red kangaroos in relation to their drought-related mortalities, rainfall and forage quality.

Key words: herbivore, kangaroo, juvenile mortality, fibre digestion, gut fill.

Introduction

Red kangaroos (*Macropus rufus* Desmarest) are large (>20 kg) herbivorous marsupials of arid and semi-arid inland Australia (Dawson, 1995). The population dynamics of red kangaroos appear tightly linked with environmental factors, mediated largely through juvenile survival and recruitment (Bayliss, 1987; Shepherd, 1987). Similar patterns are seen in a number of other large mammalian herbivores (Owen-Smith, 1990; Clutton-Brock et al., 1992; Sæther, 1997; Gaillard et al., 1998; Gaillard et al., 2000; Owen-Smith et al., 2005). Juvenile mortalities in large herbivores are usually low during favourable environmental conditions, but typically very high under poor conditions (e.g. Forchhammer et al., 2001; Clutton-Brock et al., 1992). Juvenile survival therefore is a major limiting factor for many large herbivores, irrespective of climate/habitat (e.g. temperate *vs* tropical; forest *vs* grassland) or of the proximate causes of mortality (e.g. predation, disease, malnutrition) (Sæther, 1997; Gaillard et al., 1998; Gaillard et al., 2000). This also appears to be the case for the large kangaroos in Australia (Dawson, 1995).

In arid and semi-arid Australia, rainfall is the most notable environmental factor affecting kangaroo populations (Newsome et al., 1967; Bayliss, 1985a; Bayliss, 1985b; Robertson, 1986; Shepherd, 1987; Caughley et al., 1985; Cairns and Grigg, 1993; Dawson, 1995; McCullough and McCullough, 2000). Rainfall has a strong influence on cohort survivorship and population age-structure (Robertson, 1986). Juvenile mortality is typically high during drought (Dawson, 1995; McCullough and McCullough, 2000) and only after several rain periods does juvenile survival improve, leading to significant population recruitment (Newsome et al., 1967; Robertson, 1986; Dawson, 1995). The mechanism by which

rainfall affects juvenile survival has not been defined, but recent work suggests that both diet quantity and diet quality are important (Moss and Croft, 1999; Munn and Dawson, 2003a; Munn and Dawson, 2003b; Munn et al., in press).

Adult red kangaroos are able to utilise fibrous vegetation by fermentative digestion in a large colon-like forestomach (Forbes and Tribe, 1965; Hume, 1974). Primarily grass-eaters, red kangaroos prefer young, green vegetation low in fibre (i.e. the structural carbohydrates cellulose and hemicellulose and also lignin/cutin) and high in nitrogen and easily digestible cell contents (Chippendale, 1968; Griffiths and Barker, 1966; Dawson et al., 1975; Ellis et al., 1977). During wet seasons, red kangaroos may also consume significant quantities of young forbs (non-woody dicots) (Dawson and Ellis, 1994). However, during dry seasons fresh plant growth is quickly eaten out and mature, fibrous grasses predominate (Bailey et al., 1971; Barker, 1987). Increasing fibre content is a major factor reducing the digestibility of most grasses (Burton et al., 1964; Terry and Tilley, 1964; Short et al., 1974; Ballard et al., 1990). During dry seasons, fibrous grasses provide up to 90% of the adult red kangaroo diet (Dawson and Ellis, 1994), though the extent to which smaller, juvenile red kangaroos can utilise fibrous forage is uncertain. Moreover, during prolonged or severe drought, red kangaroo mothers usually cease lactating (Frith and Sharman, 1964; Newsome, 1964a; Newsome, 1964b), making dependent young solely reliant on available forage.

Like all marsupials, red kangaroos are extremely underdeveloped at birth, weighing just 0.8 g. They spend their first 6 months of life within a large well-developed pouch, a characteristic of macropodid marsupials (Frith and Sharman, 1964; Sharman et al., 1964). By 230–250 days, they permanently exit the mother's pouch, becoming a 'young-at-foot' (mass 4-5 kg). Young-at-foot (YAF) kangaroos continue to suckle from their mother, accessing the same teat they used during pouch life. Forage intake at this stage increases markedly and red kangaroos are fully weaned at around one year (mass 10–12 kg) (Sharman et al., 1964; Dawson, 1995). It is this age/size class, from permanent-pouch-exit (i.e. YAF) until shortly after weaning, that red kangaroos appear most vulnerable to dry conditions (Shepherd, 1987; Dawson, 1995; McCullough and McCullough, 2000), when mainly fibrous vegetation is available (Dawson and Ellis, 1994).

Generally, the digestion of fibrous forage is less efficient among small herbivores (Parra, 1978; Demment and Van Soest, 1985; Illius and Gordon, 1992; Cork, 1994). Smaller animals have higher mass-specific metabolic rates than larger species, but also smaller absolute gut sizes and necessarily faster rates of food passage (Mould and Robbins, 1982; Demment and Van Soest, 1985; Illius and Gordon, 1992). Compared with larger animals, material ingested by smaller herbivores is exposed to microbial action for a shorter period, thereby reducing digestive efficiency (i.e. nutrient extracted per unit feed ingested). Furthermore, metabolic rate in mammals scales with a body-mass exponent of less than 1, often 0.75 (Kleiber, 1975; Schmidt-Nielsen, 1984; Hayssen and Lacy, 1985), but gut size scales with a body-mass exponent equal to one (Demment and Van Soest, 1985). Therefore, compared with larger species, smaller herbivores have lower gut capacities relative to their metabolic energy requirements ('metabolic-gut-capacity'). The minimum body size predicted for effective fibre digestion by foregut fermenting herbivores is ~9–15 kg (Parra, 1978; Demment and Van Soest, 1985). This is close to the weaning body mass of young kangaroos (Dawson, 1995). In addition to the potential constraints of small gut size and high mass-specific metabolism, juvenile animals are also faced with additional costs associated with growth. These costs can be substantial (Munn and Dawson, 2003a; Munn and Dawson, 2003b), further limiting the potential for young kangaroos to utilise high-fibre forage (Munn et al., in press). Here we further explore the impact of fibrous forage on digestive capabilities of YAF, weaned and mature red female kangaroos. In particular we compare the ability of juvenile and adult kangaroos to ingest and digest forages with markedly different fibre contents (neutral- and acid-detergent fibre) in relation to body size, rates of food passage and dry matter gut fill.

Materials and methods

Experimental animals

Juveniles

Six juvenile red kangaroos (four females, two males) were taken from their mothers shortly before permanent-pouch-exit. They were aged from foot and tail lengths (Sharman et al., 1964) and treated for parasites (internal and external) using Ivermectin (0.2 mg kg^{-1}; Large Animal Ivomec, Merck, Sharpe and Dohme, Granville, Australia). The young kangaroos were reared in artificial pouches (Williams and Williams, 1999) until they reached the age of permanent-pouch-exit, about 250 days (Sharman et al., 1964). Five weeks prior to experimentation the animals were transferred to our laboratory animal house and maintained in pens (430 cm×120 cm×250 cm) under a 12 h:12 h L:D cycle. The now YAF kangaroos were weighed at the beginning of each week (±0.05 kg). Rabbit Pellets (Gordon's Specialty Stock Feeds, Yanderra, Australia), Kangaroo Cubes (Doust and Rabbidge, Forbes, Australia), a lucerne/wheat bran mix (Kensington Produce, Sydney, Australia) and water were available *ad libitum*. This diet was supplemented with Digestelact (Digestelact Low Lactose, Sharpe Laboratories, Sydney), a low-lactose milk powder commonly used for hand-rearing orphaned marsupials (Williams and Williams, 1999), made to full strength (125 g 900 ml^{-1} H_2O). During non-experimental periods a daily supplement of 100 ml of Digestelact was offered to the YAF. Milk intakes by red kangaroo young have not been reported, but on this intake of Digestelact, plus forage and pellets, the YAF red kangaroos maintained growth rates comparable to those reported by Sharman et al. and Frith and Calaby (Sharman et al., 1964; Frith and Calaby, 1969).

Milk was withheld from the YAF according to the diet

treatments described below. Milk intake was reduced over time until it was eliminated at normal weaning age, ~360 days. During the rearing process, juveniles were exposed to fresh grass and soil, and to the faeces of adult red kangaroos, to facilitate infection by the microbes needed for the proper functioning of the kangaroo forestomach.

Feeding trials were carried out when the average age (± standard error of the mean; s.e.m.) of the YAF was 302±6 days and their average body mass was 6.4±0.2 kg. Trials were repeated using the same animals after they had been fully weaned and were, on average, 394±7 days and 10.9±0.3 kg body mass.

Adults

Six tame non-lactating adult female red kangaroos from a captive colony were maintained under housing conditions identical to those of the juveniles. Kangaroo Cubes, the lucerne/wheat bran mix and water were available *ad libitum*. Five of the adult females were known to be at least 5 years old; the other was 4 years old. Average body mass of the adult females during the experiments was 25.8±1.6 kg.

Diets and feeding regimens

Two forages of different fibre levels were used. Chopped lucerne (alfalfa, *Medicago sativa*) hay was considered high-quality forage, being comparatively low in neutral-detergent fibre and high in nitrogen (N). Chopped oaten (*Avena sativa*) hay was considered poor-quality forage, being higher in fibre and low in N content (Table 1). The N and fibre contents of the chopped oaten hay were similar to that of grasses foraged by red kangaroos during a severe drought in arid rangeland of western NSW (Bailey et al., 1971). Apparent dry matter digestibility of lucerne and oaten hays by adult red kangaroos was ~55% and 45%, respectively (McIntosh, 1966; Hume, 1974). Some animals initially refused the chopped oaten hay. Subsequently, the diet was always lightly sprayed (<5% v/w) with unsweetened apple juice (Golden Circle, Sydney, Australia) to increase palatability. The contribution of the juice to energy and nitrogen intakes was assessed as negligible.

In preliminary trials, YAF red kangaroos offered only chopped oaten hay (i.e. without a milk supplement) eventually did not eat. This treatment was therefore omitted from the main trials. Only by using a milk supplement (80 ml day^{-1} of full-strength Digestelact) were we able to assess the YAF capabilities when fed chopped oaten hay. Milk was offered at 09:00 h and 18:00 h at 40 ml per feed and was always completely consumed. This level of milk intake was used to mimic that likely to be available to YAF red kangaroos exposed to harsh environmental conditions. The importance of the milk supplement to forage digestion by YAF red kangaroos was described by Munn and Dawson (Munn and Dawson, 2003a).

Diet order was established by randomly allocating three YAF to a starting diet of chopped lucerne or oaten hay (with milk). The YAF were then assigned a fixed regimen of lucerne, followed by chopped oaten hay with milk or *vice versa*. After weaning, feeding trials were repeated using the same animals, maintaining the diet order initially established. Importantly, weaned red kangaroos did not receive any milk supplement. Similarly, three adult females were randomly assigned to a starting diet of chopped lucerne followed by oaten hay, the order being reversed for the other three adults. This fixed, counter-balanced design was used to minimise carry-over affects caused by diet order or animal age (Zar, 1999).

Experimental procedure

Feeding trials were conducted in a temperature-controlled room (25°C) on a 12 h:12 h L:D cycle, with lights on at 06:00 h. Adult and juvenile kangaroos were housed individually in metabolism cages (1245 mm×960 mm× 1670 mm) with mesh floors. Faeces and urine passed to a collection tray beneath each cage. Collection trays consisted of a fine wire mesh on which faeces and any spilt feed were trapped, allowing urine to flow to the tray bottom where it drained continuously into polyethylene volumetric flasks. Food and water containers were attached to the outside of each cage to minimise feed spillage. Food and water containers and waste collection trays were cleaned daily with demineralised water.

Table 1. *Composition of the chopped lucerne and oaten hays and Digestelact* milk powder*

	Lucerne hay	Oaten hay	Digestelact
Dry matter (% initial mass)	92.2±0.6	92.4±0.4	99.0±0.0
Ash (% DM)	9.6±0.3	5.8±0.1	4.84±0.0
Organic matter (% DM)	90.4±0.3	94.2±0.1	95.2±0.0
Gross energy (kJ g^{-1} DM) (kJ ml^{-1} milk)	16.7±0.2	15.7±0.4	21.9±0.2 (2.7±0.03)†
Nitrogen (% DM) (mg ml^{-1} milk)	2.9±0.07	0.89±0.09	4.5±0.01 (5.6±0.01)†
Neutral detergent fibre (% DM)	43.4±0.8	64.3±0.5	–
Acid detergent fibre (% DM)	32.4±1.0	36.0±0.3	–
Lignin/cutin (% DM)	6.2±0.2	3.4±0.1	–
Hemicellulose (% DM)	11.0±0.2	28.4±0.2	–
Cellulose (% DM)	26.2±0.8	32.5±0.2	–
Cell contents (% DM)	56.6±0.8	35.7±0.5	–

DM, dry matter; *Digestelact low lactose (Sharpe Laboratories, Sydney, Australia).
Values are means ± s.e.m. †Values in parentheses are for Digestelact at normal dilution (i.e. 124 g DM l^{-1} milk).

During the preliminary and feeding trials herbage was offered to the kangaroos at twice the previous day's level of voluntary intake.

YAF and weaned kangaroos were allowed 5 days to acclimate to their metabolism cage before a preliminary trial commenced. Preliminary trials were conducted for at least 5 days or until food intake was stable, after which time a 5-day feeding trial commenced. During each trial, food refusals and faeces were collected quantitatively and bulked over the 5 days and stored frozen. YAF and weaned kangaroos were weighed (±0.05 kg) at the same time each day throughout the preliminary and feeding trials. At the end of each trial, animals received their usual diet (i.e. water, kangaroo cubes, rabbit pellets, lucerne/bran mix) *ad libitum* for at least 10 days; YAF also received 100 ml Digestelact day^{-1}.

Adult red kangaroos were allowed at least 10 days to acclimate to their experimental diet and metabolism cage (i.e. preliminary trial). After food intake had stabilised a 5-day feeding trial commenced. Adult kangaroos were weighed (±0.05 kg) at the beginning of the preliminary and experimental trials and again at the end of the experimental trial. After each experimental trial adults received their usual diet (water, kangaroo cubes, lucerne/bran mix) *ad libitum* for at least 10 days.

Analysis of samples

Samples of food offered, together with all food refused and all faeces were collected daily and stored frozen. Foodstuffs and faeces were later thawed and sub-samples (~20% wet mass) were prepared for analysis by air-drying in a forced convection oven at 50°C (Robertson and Van Soest, 1981) for 48 h. Further sub-samples were dried at 90°C for a further 24 h, but there was no change in DM contents. Dried samples were ground through a 1 mm mesh in a Wiley Mill (Arthur Thomas Co., Scientific Apparatus, Philadelphia, USA). Ash contents of dried, ground foodstuffs (including Digestelact milk powder) and faeces were determined in duplicate by dry-ashing 0.5 g samples at 550°C overnight in a Thermolyne Muffle Furnace (Model 62700; Dubuque, Iowa, USA). Organic matter content of foods and faeces were calculated as DM – ash content.

Neutral-detergent fibre (NDF; largely cellulose, hemicellulose and lignin/cutin), acid-detergent fibre (ADF; largely cellulose and lignin/cutin) and lignin/cutin contents of feeds were determined in duplicate from 0.5 g samples using a sequential filter-bag technique and an ANKOM Fibre Analyser (Model 220, ANKOM Technology Corp., Fairport New York, USA). The reagents and general procedures used were as described by Van Soest et al. (Van Soest et al., 1991). Prior to neutral-detergent digestion, samples were treated with 1 ml of heat stable α-amylase (Sigma A – 3306; Sigma Aldrich Pty Ltd, Sydney) for 80 min to remove starch (Van Soest et al., 1991). Sodium sulphite and decalin were removed from the neutral-detergent procedure (Robertson and Van Soest, 1981; Van Soest et al., 1991). This sequential filter-bag technique ensured no unintentional loss of sample throughout. Soluble cell contents (DM–NDF), and contents of hemicellulose (NDF–ADF) and cellulose (ADF–lignin/cutin) in foods and faeces were determined by difference.

Energy contents of dried ground foodstuffs were determined by combusting duplicate sub-samples of 0.7 g in a ballistic bomb calorimeter (Gallenkamp, Model CB-375; Gallenkamp and Co. Ltd, UK), using a benzoic acid standard for calibration every 25 samples. The total nitrogen content of foodstuffs was determined in duplicate by total combustion of approximately 0.2 g samples in a Leco CHN-1000 elemental analyser (Leco Inc. St Joseph, Michigan, USA).

Food intake and apparent digestibility

Some kangaroos showed considerable diet selection during the feeding trials. Intake of dietary components (e.g. DM) was therefore calculated as:

$$\text{Intake} = (\text{component in FO} \times \text{mass FO}) - (\text{component in FR} \times \text{mass FR}) , \quad (1)$$

where FO=food offered, FR=food refused, and all masses are in g. Apparent digestibility (%) of dietary components was then calculated as:

$$[(\text{Intake} - \text{Faecal output}) / \text{Intake}] \times 100 , \quad (2)$$

where intake and faecal output are in g day^{-1} (Robbins, 1993).

Food passage and mean retention times

The rate of passage of solutes and particles through the gastrointestinal tract of kangaroos was measured using two inert markers. The solute marker used was cobalt-ethylene diaminetetraacetic acid (Co-EDTA), prepared according to Udén et al. (Udén et al., 1980). Particles were marked with chromium mordanted to cell walls (Cr-CW) according to Udén et al. (Udén et al., 1980); mordanting renders particles indigestible. Cell walls were prepared from chopped lucerne hay (dried at 50°C and ground through a 1 mm mesh) by the neutral detergent method (Robertson and Van Soest, 1981; Van Soest et al., 1991) and wet-sieved through a series of Endicott (London, England) screens. Particles that passed through a 1-mm screen but were trapped on a 600 μm screen were retained for mordanting.

Doses of Co-EDTA and Cr-CW were mixed, lightly sprayed with unsweetened apple juice and offered to the YAF, weaned and adult red kangaroos on day 2 of each feeding trial at 11:00 h. Some adults initially refused the marked food, which was subsequently spread on one half of a slice of 3-day-old fruit bread (Buttercup Bakeries, Moorebank, NSW, Australia), which was readily consumed. YAF and weaned kangaroos were offered 1.0 g Cr-CW and 0.5 g Co-EDTA. Adults were offered 2.0 g Cr-CW and 1.0 g Co-EDTA. Six mature female and juvenile kangaroos were offered the marked dose during each feeding trial, but only five animals from each age class completely consumed the marked foods on each occasion. In these cases, marked feeds were consumed within 5 min, and thus were considered a pulse dose. Exact doses, however, were unknown because a small amount of marker often remained in the bowl, and some was lost with spilt saliva. After dosing, faeces were collected from the YAF and weaned kangaroos at

4 h intervals for 24 h, followed by 6 h intervals for 24 h, then 8 h intervals for 24 h and 12 h intervals for a further 24 h (total 96 h). After dosing, faecal samples were collected from the adult kangaroos at 6 h intervals for 48 h, then 8 h intervals for 24 h and 12 h intervals for a further 24 h (total 96 h).

Faecal samples were analysed for Co and Cr concentration using an Inductively Coupled Plasma Optical Emission Spectrometer (ICP OES, Optima 3000DV, Perkin Elmer, CT USA). Dry faecal samples (~0.5 g) were prepared for ICP analysis according to Mambrini (Mambrini, 1990), but see also Caton et al. (Caton et al., 1996). Samples were dry-ashed overnight at 550°C in a Thermolyne Muffle Furnace (Model 62700; Dubuque, Iowa, USA). Ash residue was then boiled for 3 min in 10 ml of 2% HNO_3 containing 2 g l^{-1} CaCl (matrix solution). After boiling, solution and residue were completely transferred to 25 ml volumetric flasks, allowed to cool and made up to the mark with the matrix solution. The resultant solutions were allowed to stand for 24 h before the supernatant was carefully drawn off for analysis. Standards for ICP were prepared using Co-EDTA and Cr-dichromate dissolved in the matrix solution.

Mean retention times (MRT) for solute and particle markers through the entire gastrointestinal tract (Warner, 1981) were calculated according to Blaxter et al. (Blaxter et al., 1956):

$$\mathrm{MRT} = \sum_{i=1}^{n} M_i T_i \Big/ \sum_{i=1}^{n} M_i , \qquad (3)$$

where MRT is in h, M_i is the amount of marker excreted in the ith defecation at time T_i (equal to the time elapsed from dosing to the mid-point of each faecal collection period) and n is the total number of defecations in which marker could be detected. MRT is the single most useful measure of rate of passage (Warner, 1981).

An index of total gastrointestinal DM content (i.e. dry gut fill) was calculated from faecal output, particle MRT and apparent dry matter digestibility (Holleman and White, 1989). Indigestible fill (V_N; g DM) was calculated as:

$$V_N = F \times \mathrm{MRT} , \qquad (4)$$

where F=faecal output (g DM h^{-1}) and MRT=particle MRT (h). Total digestive tract fill (V; g DM), was then calculated as the sum of the indigestible and digestible fill according to:

$$V = V_N + \left[\frac{V_N A}{2(1-A)} \right] , \qquad (5)$$

where A=apparent DM digestibility. It should be noted that the marked particles used to measure particle MRT may not have represented the full range of digesta particle sizes, but our results provide a useful index of total DM digesta contents (see also Gross et al., 1996).

Statistical analysis

Although the method of choice for statistical comparisons was analysis of covariance (ANCOVA) with body mass as the covariate, the YAF and the weaned kangaroos were the same animals, and hence the data sets were not independent. Also, it was not logistically feasible to include greater numbers of such large animals in the study. These constraints meant that the use of ANCOVA resulted in overly complex comparisons with small numbers of replicates and limited statistical power. Instead, repeated-measures analysis of variance (RM-ANOVA) was used to compare within and between group data from YAF, weaned and adult red kangaroos. As noted, non-independence of the YAF and weaned kangaroos prevented their combined analysis with adult data (Zar, 1999). Therefore, YAF and weaned kangaroos were compared using two-way RM-ANOVA with two levels of within-group factors (diet and age). YAF and weaned data were then compared separately to those of adult red kangaroos using two-way RM-ANOVA. Statistical outcomes for the within-YAF, within-weaned and within-adult kangaroo data were the same across all between-group comparisons (e.g. outcomes for within-YAF data were the same when YAF were compared to weaned or adult kangaroos). For this reason results are presented as if they were one data set, even though they were tested independently.

Data on intake and output are presented as g day^{-1}, or as g $kg^{-0.75}$ day^{-1} for comparison with other studies. The most appropriate exponent for intra-specific comparisons often differs from 0.75 (Hume, 1999), but there were insufficient data to establish this relationship in this study. Therefore we used a body-mass exponent of 0.75, which was shown (Hayssen and Lacy, 1985) to be the most appropriate for comparisons of basal metabolic rate among marsupials.

Assumptions for ANOVA were tested using the Kolmogorov–Smirnov test for normality (α=0.05) and Levene's test for homogeneity of variances (α=0.05). To achieve normality and or homogeneity, log (+1) transformations were applied to the following data sets: DMI (g day^{-1}; YAF *vs* adults), digestible DMI (g day^{-1}; YAF *vs* weaned, YAF *vs* adults), organic matter intake (OMI; g day^{-1}; YAF *vs* adults) and digestible OMI (g day^{-1}; YAF *vs* weaned, YAF *vs* adults), gut fill (g DM) (YAF *vs* weaned). Some data sets, however, could not be normalised and were compared using a Friedman's test (a non-parametric ANOVA for repeated measures) (Zar, 1999). Data sets compared using Friedman's test included: apparent digestibility (%) of DM, organic matter (OM), soluble cell contents (YAF *vs* adult, YAF *vs* weaned), digestible DMI (g $kg^{-0.75}$ day^{-1}; YAF *vs* weaned) and digestible OMI (g $kg^{-0.75}$ day^{-1}; YAF *vs* weaned). Proportional data for all digestibilities were arcsine transformed (Zar, 1999).

Significant differences detected by ANOVA were further investigated using a Tukey's Honest Significant Differences (HSD) *post hoc* test. Significant differences detected by Friedman's test were investigated using equation 11.3 from Zar (Zar, 1999) with standard error adjusted for repeated measures (Zar, 1999; Equation 12.53). ANOVA, Tukey HSD and Friedman's tests were performed using Minitab for Windows 12.1 (1998; Minitab Inc., PA, USA).

Results

Forage intake and apparent digestion

Although the YAF red kangaroos were just 25% of the mature, non-reproductive female's body mass (Table 2) they ingested 56% as much DM (g day^{-1}) from the high-quality chopped lucerne hay as did the mature females (Table 3). By weaning age, DMI (g day^{-1}) by the juvenile red kangaroos had increased to a level that was not significantly different from that of adults, despite these younger animals being less than half the body mass of mature females (Table 2). Apparent DM digestibility of chopped lucerne hay was similar across all of the age classes (55–59%) (Table 3). Consequently, patterns of digestible DMI by the red kangaroos on chopped lucerne hay largely followed those for gross DMI (Table 3). On an allometric basis (i.e. per kg$^{-0.75}$), however, digestible DMI by the YAF and weaned kangaroos on chopped lucerne hay was 1.5–1.7 times that by mature females, but they were not significantly different from each other (Table 3).

On the poor-quality chopped oaten hay, gross DMI (g day^{-1}) by the mature female kangaroos was not significantly lower than that on chopped lucerne (Table 3). This was not the case, however, for the juvenile red kangaroos, which had DMIs (g day^{-1}) of chopped oaten hay that were less than half that of chopped lucerne (P<0.01; Table 3). The apparent digestibly of DM from the oaten hay diet was similar across all age classes (43–45%; P>0.05) (Table 3). However, it must be remembered that the YAF animals on this diet also received a small portion of artificial milk (80 ml day^{-1}). Preliminary trials indicated that the YAF would not have survived on chopped oaten hay alone (see Munn and Dawson, 2003a). Assuming a milk–DM digestibility of 95% (Penning et al., 1977; Roy, 1980; Ternouth et al., 1985), the apparent digestibility of DM from the chopped oaten hay alone (i.e. excluding milk) by the YAF kangaroos was just 34.2±3.1%, significantly lower than that by weaned or adult kangaroos (Table 3).

Table 2. *Changes in body mass over 5 days by young-at-foot, weaned and adult red kangaroos fed chopped lucerne and oaten hays*

	YAF	Weaned	Adult
Initial body mass (kg)			
Lucerne hay	6.4±0.5^{Z}	10.8±0.3^{Y}	25.8±1.5^{X}
Oaten hay	6.1±0.3^{Z}	10.9±0.5^{Y}	25.7±1.6^{X}
Mass change (g day^{-1})			
Lucerne hay	43±9^{X}***	55±11^{X}***	0.0±16^{X}
Oaten hay	–32±14^{X}	–33±14^{X}	–61±30^{X}
Mass change (% initial body mass)			
Lucerne hay	3.3±0.67^{A}***	2.6±0.47^{A}**	0.1±0.31^{B}
Oaten hay	–2.3±0.96^{A}	–1.5±0.65^{A}	–1.2±0.54^{A}

YAF, young-at-foot.

Values are means ±s.e.m. (N=6).

Within a row, values with different superscript letters are significantly different ($^{A,B,C}P$<0.05; $^{X,Y,Z}P$<0.01). Asterisks indicate differences between diets within ages (**P<0.01 and ***P<0.001).

Fibre digestion

The digestibility of soluble cell contents by juvenile and adult red kangaroos fed chopped lucerne hay was high (75–79%) and not significantly different between age groups (Table 4). Differences were apparent, however, in their ability to digest forage fibre. The YAF kangaroos digested significantly less NDF, ADF, cellulose and hemicellulose than did the mature female kangaroos (Table 4). YAF also digested significantly less NDF, ADF and cellulose (but not hemicellulose) than the weaned juveniles (Table 4).

On the higher fibre forage of chopped oaten hay, the digestibility of soluble cell contents by YAF, weaned and mature female kangaroos was significantly lower than on chopped lucerne (Table 4). The YAF kangaroos also digested significantly less soluble cell contents from oaten hay than did weaned or adult kangaroos, by ~5–7 percentage units. The digestibility of NDF from chopped oaten hay was also lower in all age groups, by around 10 precentage units, compared with chopped lucerne. These differences were greatest for the YAF kangaroos, which digested significantly less NDF, ADF, cellulose and hemicellulose than either the weaned or mature females (Table 4). The pattern of fibre digestibility by weaned juveniles was generally intermediate between the YAF and adult kangaroos (Table 4).

Digesta mean retention times and gastrointestinal DM contents (DM gut fill)

Cumulative elimination curves describing the pattern of solute and particulate passage through the gastrointestinal tract of our kangaroos are outlined in Fig. 1. Solute and particle markers were completely eliminated from the digestive tract of the YAF, weaned and adult kangaroos by 60 h and 96 h post dose, respectively. There was significant separation of solute and particle digesta markers in the YAF, weaned and mature female kangaroos on both chopped lucerne and oaten hays, with the solute marker universally eliminated faster than the particle marker (Fig. 1; Table 5; P<0.01). Overall, the juvenile red kangaroos showed the greatest response in their patterns of solute and particle elimination in relation to diet.

Differences in the elimination patterns of particle and solute markers between the juvenile and mature female kangaroos were apparent when we considered overall MRTs. On high-quality, chopped lucerne hay, the MRTs for the solute marker were significantly shorter in the YAF and weaned kangaroos than in adults, by ~4 h. Particle marker MRTs on chopped lucerne, however, were not significantly different between age groups (Table 5). However, on the more fibrous chopped oaten hay, MRTs for solutes and particles were significantly longer than on the chopped lucerne in both the YAF and weaned kangaroos; the solute marker by ~6 h and the particle marker by ~10 h in each case. Conversely, MRTs for solute and particle markers through mature females were not significantly affected by diet (Table 5).

As expected, indices of gastrointestinal DM contents (i.e. DM gut fill; in g) on chopped lucerne and oaten hays were lowest in the YAF red kangaroos, followed by weaned and then

Table 3. *Intakes and digestion of chopped lucerne and oaten hays by young-at-foot, weaned and adult red kangaroos*

	YAF	Weaned	Adult
DMI (g day^{-1})			
Lucerne hay	233±23^{B}** (**)	370±14^{A}***	414±38^{A}
Oaten hay (from forage†)	105±10^{C} (95±10^{C})	176±23^{B}	345±47^{A}
Apparent DM digestibility (%)			
Lucerne hay	55.8±0.6^{A}*** (***)	59.3±1.4^{A}***	56.9±2.8^{A}***
Oaten hay (from forage†)	40.3±3.4^{A} (34.2±3.1^{B})	42.0±2.9^{A}	41.8±1.1^{A}
Digestible DMI (g day^{-1})			
Lucerne hay	130±14^{Y}*** (***)	219±6^{X}***	231±14^{X}
Oaten hay (from forage†)	40±2^{Z} (31±1^{Z})	71±6^{Y}	144±20^{X}
Digestible DMI (g kg$^{-0.75}$ day^{-1})			
Lucerne hay	32.1±1.7^{B}*** (***)	36.8±0.6^{B}***	20.5±1.7^{A}**
Oaten hay (from forage†)	10.4±0.2^{A} (8.0±0.2^{B})	11.9±0.9^{A}	12.8±1.8^{A}
OMI (g day^{-1})			
Lucerne hay	210±20^{B}** (***)	330±15^{A}***	374±34^{A}
Oaten hay (from forage†)	100±11^{C} (90±11^{C})	165±22^{B}	332±45^{A}
Apparent OM digestibility (%)			
Lucerne hay	54.9±0.7^{A}*** (***)	58.7±1.1^{A}***	57.2±3^{A}***
Oaten hay (from forage)	42.2±2.6^{A} (35.9±2.3^{B})	43.4±2.8^{A}	44.6±1^{A}
Digestible OMI (g day^{-1})			
Lucerne hay	116±12^{Y}*** (***)	193±6^{X}***	209±13^{X}
Oaten hay (from forage)	41±2^{Z} (32±2^{Z})	69±7^{Y}	147±20^{X}
Digestible OMI (g kg$^{-0.75}$ day^{-1})			
Lucerne hay	28.4±1.5^{Y}*** (***)	32.5±0.5^{Y}***	18.6±1.6^{X}**
Oaten hay (from forage)	10.5±0.4^{A} (8.0±0.4^{B})	11.6±0.9^{A}	13.1±1.8^{A}

YAF, young-at-foot; DM, dry matter; DMI, dry matter intake; OM, organic matter; OMI, organic matter intake.

Values are means ±s.e.m. (N=6).

Within a row, values with different superscript letters are significantly different ($^{A,B,C}P<0.05$; $^{X,Z}P<0.01$). Asterisks indicate differences between diets within ages (**P<0.01 and ***P<0.001). †Apparent digestibility and digestible intakes of DM and OM from forage were estimated assuming a milk digestibility (DM and OM) of 95% (see text). Asterisks in parentheses in the YAF column indicate significant differences between lucerne hay and oaten hay forage after accounting for the 95% digestibility of the milk supplement (**P<0.01 and ***P<0.001).

mature females (Table 6). However, there was considerable variation around the mean in both the YAF and weaned kangaroos, and differences in DM gut fill between the younger ages were not significant, though weaned animals tended to have greater fills on both diets at P=0.09 (Table 6). When offered chopped lucerne hay, the YAF kangaroos had the highest mass-specific DM gut fills (i.e. g kg^{-1}), significantly higher than that of mature females (P<0.05) and tending toward significance when compared with weaned animals (P=0.07). When offered chopped oaten hay, however, there were no significant differences in mass-specific DM gut fills of YAF, weaned or mature female kangaroos. In other words, on high-quality chopped lucerne, the mature female kangaroos had mass-specific DM gut fills that were just 65% of that seen in the YAF, but significantly increased DM gut fill on chopped oaten hay by around 1.6-fold. Conversely, mass-specific DM gut fill in YAF and weaned kangaroos was not significantly affected by diet (Table 6). On an allometric basis (i.e. g DM kg$^{-0.75}$), there were no significant differences in DM gut fill among the YAF, weaned and mature females on chopped lucerne hay, though variation was high, particularly in the juveniles (Table 6). On chopped oaten hay, allometrically related DM gut fill in mature females was not significantly different from that of weaned and YAF kangaroos (P=0.15 and P=0.11, respectively), but was significantly greater than that of adults fed chopped lucerne (Table 6).

Discussion

Adult red kangaroos are highly sexually dimorphic and this is reflected in their levels of feed intake on good-quality forage (e.g. lucerne hay) (Munn et al., in press). The mature females in our study were older than four years and had body masses of 25–30 kg. Mature males, on the other hand, can attain average body masses of 60–80 kg by 15 years of age (Dawson, 1995). Our mature females were not lactating and ingested half as much chopped lucerne hay (414 g DM day^{-1}; Table 3) as did mature male red kangaroos (823 g DM day^{-1}; N=3, body

Table 4. *Digestibility (%) of cell contents and forage fibre components by young-at-foot, weaned and adult red kangaroos fed chopped lucerne and oaten hays*

	YAF	Weaned	Adult
Cell contents (%)			
Lucerne hay	75.5 ± 0.3^{A}***	75.9 ± 0.5^{A}***	73.4 ± 0.8^{A}***
Oaten hay	61.6 ± 1.9^{B}	69.0 ± 1.7^{A}	67.1 ± 1.6^{A}
NDF (%)			
Lucerne hay	27.3 ± 1.4^{Z}***	33.7 ± 0.8^{Y}***	40.4 ± 2.3^{X}***
Oaten hay	18.2 ± 1.9^{Z}	23.4 ± 2.2^{Y}	28.6 ± 2.0^{X}
ADF (%)			
Lucerne hay	21.8 ± 1.8^{Z}***	29.5 ± 1.5^{Y}***	35.3 ± 2.8^{X}***
Oaten hay	14.5 ± 1.8^{Z}	17.7 ± 2.4^{Y}	28.1 ± 1.9^{X}
Cellulose (%)			
Lucerne hay	29.7 ± 2.8^{Z}***	36.2 ± 1.4^{Y}***	42.5 ± 3.0^{X}***
Oaten hay	14.1 ± 1.4^{Z}	19.4 ± 2.4^{Y}	23.7 ± 3.4^{X}
Hemicellulose (%)			
Lucerne hay	41.1 ± 3.2^{Y}***	43.0 ± 3.2^{Y}**	53.2 ± 2.1^{X}***
Oaten hay	22.3 ± 2.8^{Y}	26.9 ± 1.9^{X}	28.6 ± 2.7^{X}

YAF, young-at-foot; NDF, neutral-detergent fibre; ADF, acid-detergent fibre.

Values are means ±s.e.m. (N=6).

Within a row, values with different superscripts are significantly different ($^{A,B,C}P<0.05$; $^{X,Y,Z}P<0.01$). Asterisks indicate differences between diets within ages (**$P<0.01$ and ***$P<0.001$).

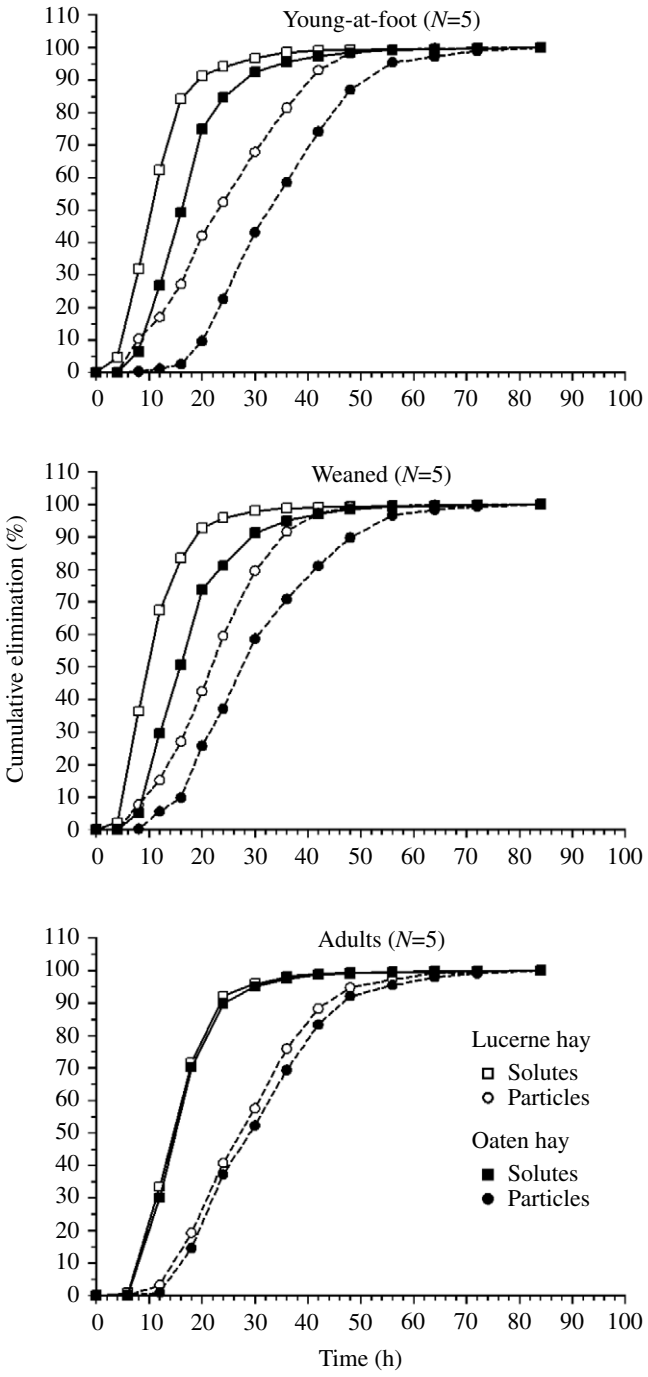

Fig. 1. Mean cumulative elimination (or appearance) of the solute (Co) and particle (Cr) markers in faeces (% total marker eliminated) from young-at-foot (YAF), weaned and adult red kangaroos fed chopped lucerne (open symbols) and oaten hays (closed symbols) following a pulse dose at time (h) zero.

mass=62±5 kg; McIntosh, 1966). Apparent digestibility of DM from chopped lucerne hay by our females was identical to that by McIntosh's males, averaging 56% (McIntosh, 1966). Thus, on an allometric basis (i.e. per $kg^{0.75}$), digestible DMI by our mature female kangaroos on chopped lucerne hay was the same as that by mature males (McIntosh, 1966), at 21±2 g $kg^{-0.75}$ day^{-1} (Table 3). Hume reported higher intakes of chopped lucerne hay by smaller, younger male red kangaroos (29 g $kg^{-0.75}$ day^{-1}; N=3, body mass 27–33 kg) (Hume, 1974), probably because these still-growing males required additional energy and nutrients for growth. Munn and Dawson showed that, in red kangaroos, growth energy requirements have a significant impact on overall food intake (Munn and Dawson, 2003b). The YAF and weaned red kangaroos in this study were growing rapidly and had digestible DMIs (g $kg^{-0.75}$ day^{-1}) on chopped lucerne hay significantly greater than that by mature, non-lactating females (Table 3), and similar to or slightly greater than that of Hume's young red kangaroos (Hume, 1974).

On the high-quality chopped lucerne hay, the YAF and weaned red kangaroos were able to sustain growth at levels comparable with those reported by Sharman et al. for ideal conditions (Sharman et al., 1964). YAF red kangaroos at this stage normally would be taking some milk from their mothers. Our results indicate that provided sufficient high-quality forage is available, YAF red kangaroos can subsist on forage alone, at least when their energy requirements for thermoregulation are minimal. Juveniles may not be as successful under cold conditions because of their higher energy costs for thermoregulation relative to larger adults (Munn and Dawson, 2001). Cold conditions and poor-quality forage together are likely to be particularly taxing for the smaller juvenile kangaroos. YAF kangaroos on the chopped oaten hay did not

Table 5. *Mean retention times for particle (Cr) and solute (Co) markers by young-at-foot, weaned and adult red kangaroos fed chopped lucerne and oaten hays following a pulse dose*

	Mean retention time (MRT) (h)	
	Lucerne hay	Oaten hay
YAF		
Particles	24.8 ± 4.0^{A}**	34.5 ± 3.8^{A}
Solutes	11.8 ± 1.5^{X}**	17.6 ± 1.0^{X}
Weaned		
Particles	22.3 ± 3.3^{A}*	30.1 ± 4.3^{A}
Solutes	11.3 ± 1.2^{X}***	17.8 ± 1.4^{X}
Adult		
Particles	28.6 ± 2.8^{A}	30.9 ± 1.9^{A}
Solutes	15.7 ± 1.3^{Y}	16.3 ± 1.2^{X}

YAF, young-at-foot.

Values are means ±s.e.m. (N=5).

Within a row, values with different superscripts are significantly different within diets between ages (Particles $^{A,B}P<0.05$; Solutes $^{X,Y}P<0.05$). Asterisks associated with lucerne hay diets indicate significant differences between diets within ages (*$P<0.05$, **$P<0.01$ and ***$P<0.001$).

Note: MRTs were significantly longer for the particle marker than for the solute marker for all ages groups on both lucerne and oaten hay; $P<0.01$.

Table 6. *Total dry matter gastrointestinal tract fill (gut fill) in young-at-foot, weaned and adult red kangaroos fed chopped lucerne and oaten hays*

	YAF	Weaned	Adult
Gut fill (g DM)			
Lucerne hay	121.6 ± 20.8^{B}	158.6 ± 23.6^{B}	264.0 ± 23.7^{A}*
Oaten hay	113.9 ± 22.9^{B}	150.7 ± 43.3^{B}	426.5 ± 25.5^{A}
Gut fill (g DM kg^{-1})			
Lucerne hay	18.4 ± 2.8^{B}	$14.8\pm2.0^{A,B}$	10.6 ± 1.5^{A}*
Oaten hay	18.0 ± 3.4^{A}	13.5 ± 3.2^{A}	16.8 ± 0.8^{A}
Gut fill (g DM $kg^{-0.75}$)			
Lucerne hay	29.4 ± 4.5^{A}	26.7 ± 3.7^{A}	23.7 ± 2.8^{A}*
Oaten hay	28.5 ± 5.4^{A}	24.6 ± 6.2^{A}	37.7 ± 1.5^{A}

YAF, young-at-foot; DM, dry matter.

Values are means ±s.e.m. (N=5).

Within a row, values with different superscripts are significantly different within diets between ages ($^{A,B,C}P<0.05$). Asterisks associated with lucerne hay diets indicate significant differences between diets within ages ($P<0.01$).

sustain growth rates. They averaged losses of 2.3% of their body mass over 5 days, even when receiving a small milk supplement (Table 2). Loss of body condition by the YAF was partially explained by a poor digestibility of the DM of the oaten hay, leading to lower digestible DMIs (g $kg^{-0.75}$ day^{-1}) compared with weaned and adult female kangaroos (Table 3).

The poor digestibility of DM by YAF red kangaroos fed chopped oaten hay forage (i.e. after assuming a milk-DM digestibility of 95%) was partially associated with their lower digestibility of cell contents compared with weaned and adult kangaroos (Table 4). Digestibilities of cell contents by our red kangaroos (range: 60–79%) were lower than those reported for medium sized males (~88%; Hume, 1974), most probably because of differences in forage age and preparation. In juvenile and adult kangaroos the digestibility of cell contents was lower on chopped oaten hay than on lucerne (Table 4). This may be particularly important for juvenile and adult kangaroos during drought conditions, as dry, senescent grasses predominate (Dawson and Ellis, 1994). Ballard et al. found that the *in vitro* digestibility of cell contents (mainly soluble carbohydrates) declined with increasing plant age in ryegrass (*Lolium rigidium*) (Ballard et al., 1990). Also, as the proportion of soluble carbohydrates declines with maturity in grasses, the relative content of hard-to-digest structural carbohydrates (i.e. cellulose and hemicellulose) increases, thereby reducing overall dry matter digestibility (Short et al., 1974; Ballard et al., 1990).

The digestibilities of forage fibre fractions (NDF, ADF) by our mature female kangaroos were comparable to those reported for similarly sized mature males on similar forage (Hume, 1974). The smaller, juvenile kangaroos were less capable of digesting fibre fractions than were the adults. Even on the chopped lucerne hay, the YAF kangaroos digested significantly less NDF than did weaned or adult kangaroos (Table 4). Overall, fibre digestion improved with increasing body mass from YAF to weaned and adult kangaroos (Table 4). Similar patterns in fibre digestion are seen between herbivore species of markedly different body sizes. For example, Mould and Robbins found that the digestibility of NDF from chopped lucerne hay by white-tailed deer (*Odocoileus virginianus*, body mass=36 kg) was 10–15% lower that that by elk (*Cervus elaphus nelsoni*, body mass=187 kg) (Mould and Robbins, 1982). Our YAF red kangaroos were, on average, four times smaller than our adult females, but could a small body size alone explain their lower performance on the higher fibre forage, or are other factors involved?

It could be argued that, compared with weaned juveniles and adult kangaroos, the digestive system of the YAF was poorly developed by this stage. However, Norton and Dawson (M. A. Norton and T. J. Dawson, unpublished data) found that the capacity of the foregut in red kangaroos, the main site for fermentative digestion in macropodids, scaled isometrically with body mass across a wide range of age/size classes (N=22; body mass range 7–31 kg). Consequently, the fermentative capacity of the YAF foregut should be no different from that expected for an adult kangaroo of a similar body mass, assuming the microbial ecosystem of the juvenile foregut is comparable with that of adults. Griffiths and Barton showed that chemically, histologically and enzymatically the juvenile foregut in red kangaroos was comparable with that of adults by the time they reached permanent-pouch-exit (Griffiths and

Barton, 1966). By this stage, young kangaroos are ingesting significant quantities of herbage, and milk intakes are declining rapidly (Griffiths and Barton, 1966; Dawson, 1995). Similar ontological changes are seen in young ruminants during weaning. The rumen of young lambs, for example, is functionally equivalent to that of adults by 20 days after peak lactation [i.e. after they begin weaning; see Langer (Langer, 1994)]. Furthermore, by the time of final weaning our juvenile kangaroos were able to ingest as much digestible DM (g $kg^{-0.75}$ day^{-1}) from chopped oaten hay as were adult females (Table 3). Munn and Dawson, however, found that, on an allometric basis, weaned kangaroos had total daily energy requirements (kJ $kg^{-0.75}$ day^{-1}) for maintenance plus growth that were some 1.8 times that of adult females (Munn and Dawson, 2003b). Therefore, although the weaned kangaroos were able to digest chopped oaten hay DM with as much efficacy as that of adults (Table 3), they were unable to process enough of this poor-quality forage to meet their proportionally higher nutrient requirements.

The volume of forage that an animal can process depends largely on the refractory properties of the digesta and the amount of time it spends in the gut (Robbins, 1993). This is usually different for solutes (e.g. cell contents) and particles (e.g. fibrous components). Kangaroos and their relatives show digesta retention patterns typical of most non-ruminant vertebrates (Dellow, 1982; Stevens and Hume, 1995), where solutes pass through the gastrointestinal tract more rapidly than particles (Fig. 1). Notably, this is the first study in which MRTs have been measured in kangaroos using a mordanted marker for particles. Previous studies have used either stained hay particles (which lack precision) or the particle-associated marker ruthenium–phenanthroline, which migrates from larger to smaller particles during digestion (see p. 236 in Hume, 1999). Digesta separation in adult kangaroos occurs in the foregut (Dellow, 1982), where larger, fibrous particles are fermented more slowly. A typical response of red kangaroos to increasing dietary fibre content is an increase in the time that particles remain in the foregut and, consequently, a reduction in food intake (Foot and Romberg, 1965; McIntosh, 1966; Forbes and Tribe, 1970). Our adult red kangaroos, however, did not show comparable reductions in food intake when switched from low-fibre chopped lucerne to the high-fibre chopped oaten hay (Table 3), and there were also no concurrent changes in the MRT for particles or solutes (Table 5). Conversely, MRTs for solute and particle markers were significantly greater in both the YAF and weaned kangaroos when they switched from low- to high-fibre forage (Table 5), leading to significant reductions in DMI (g day^{-1}) of 52–55% (Table 3). This suggests that for adult kangaroos, adjustments in gut fill, rather than digesta retention times, may be an important response to changes in forage quality. Juvenile kangaroos, however, appeared to be at or near maximal gut fill (at least for DM) regardless of diet quality. Although we could not measure this directly, the increased MRTs in juveniles on the poor-quality oaten hay strongly suggests that they were at their limits for gut fill (g DM) on this diet (Table 6).

Our results are consistent with foraging models that indicate that metabolic-gut-capacity (i.e. gut capacity relative to metabolic rate) increases with increasing body mass (Parra, 1978; Demment and Van Soest, 1985; Illius and Gordon, 1992). Also, we have shown that intra-specifically, larger animals are capable of greater flexibility in gut fill compared with younger, smaller animals. When offered low-fibre chopped lucerne hay, mass-specific DM gut fill (g kg^{-1}) in the adult red kangaroos was just 57% of that of the YAF (Table 6). Only when fed poor-quality oaten hay did the adult female kangaroos increase mass-specific DM gut fill (g kg^{-1}) to levels comparable to that of juveniles (Table 6). Increasing gut fill allowed the mature females to maintain DMI of chopped oaten hay at levels not significantly different from chopped lucerne (Table 2). Furthermore, when the adult kangaroos were switched from lucerne to oaten hay they increased DM gut fill relative to their metabolic body mass (i.e. $kg^{-0.75}$) some 1.6 times (Table 6). Thus, when fed high-quality forage the larger, mature female kangaroos were able to reduce DM gut fill, presumably as a consequence of their lower metabolic energy requirements (Munn and Dawson, 2001; Munn and Dawson, 2003b). Because the smaller juvenile kangaroos appeared unable to compensate for lower forage quality by increasing gut fill, they could not extract sufficient nutrients from the chopped oaten hay to maintain growth (Table 2).

The age/body size at which red kangaroos may be able to relax DM gut fill on higher quality forage is unknown, but may occur around sexual maturity, at least in females. The apparent ‘reserve gut fill’ available to the mature females feeding on high-quality forage might be important for allowing the higher levels of food intake necessary to sustain peak lactation. The energy costs associated with supporting an offspring close to permanent-pouch-exit (i.e. the period of peak lactation) can be as much as 50% of the mother’s maintenance requirements (Prince, 1976), presumably necessitating higher food intakes. Increases in DM gut fill in relation to the increased energy requirements for lactation have been reported for a range of ungulate species (see Gross et al., 1996 and references therein). Red kangaroos usually cease lactating during severe or prolonged drought (Frith and Sharman, 1964; Newsome, 1964a; Newsome, 1964b), which may be related to maximal gut fill on forages insufficient to supply the nutrients needed for milk production. It would be interesting to measure how milk production changes with diet quality and gut fill in this species, particularly as DM gut fill in our mature red kangaroos was considerably less than that seen in ruminants (range 2 to >4% body mass) (see Gross et al., 1996), averaging just 1.7 % body mass on chopped oaten hay. However, that the MRTs for solute or particle markers were not increased in the mature females on chopped oaten hay suggests that they were not at their maximal gut fill on this diet.

The flexibility in gut fill in our mature female kangaroos could be due to physical expansibility of the gut or to gut hypertrophy, which is seen in other mammals and particularly those inhabiting highly seasonal environments (e.g. Weckerly, 1989; Jenks et al., 1994; Hume et al., 2002). However, in light

of the ecology of red kangaroos, expansibility of the gut may be more important than hypertrophy for ameliorating the problems associated with stochastic food supplies and quality. Because of the pressures associated with continually breeding in good times (i.e. full gut) and of the extra gut reserve needed to satisfy their own nutritional needs in bad times, there may be little selective pressure for rapid gut re-modelling in the mature female kangaroos.

To the best of our knowledge, this is the first study to show a large marsupial herbivore adjusting gut fill in response to diet quality. Plasticity in gut fill has important consequences for models of optimal foraging and digestion. These models generally assume that maximal digesta loads are directly proportional to body mass and are also the main determinant of the cessation of food intake (Demment and Van Soest, 1985; Illius and Gordon, 1992; Cork, 1994; Yearsley et al., 2001). The regulation of food intake by our mature kangaroos on the high-quality forage, however, was apparently related to factors other than the physical stimulus of gut distension. Current models using maximal digesta load as a determinant of food intake do not address situations when gut fill is not maximised. This may be particularly important when considering the foraging strategies of red kangaroos in light of their highly unpredictable environment and temporally patchy resources. Another assumption in many herbivore studies is that animals forage to maximise food (energy) intake (for a review, see Bergman et al., 2001). However, that our adult red kangaroos were not maximising DM gut fill on the high-quality lucerne hay suggests that they may not have been maximising food (energy) intake on this diet. More likely, our mature, non-lactating females were feeding to satisfy energy requirements rather than maximise intake (van Gils et al., 2003; Hume, 2005); the energy requirements of mature female red kangaroos being proportionally lower than those of still-growing juveniles (Munn and Dawson, 2003b).

Conclusion

Our data support field studies indicating that juvenile red kangaroos are limited in terms of condition and growth mainly by the availability of high-quality forage (Watson and Dawson, 1993; Dawson, 1995; Moss and Croft, 1999). As forage fibre levels increased to 40–50% NDF, the juvenile kangaroos could not sustain growth, being limited by gut capacity and higher energy requirements than adults. On chopped oaten hay, juvenile kangaroos also suffered higher nitrogen losses compared with adults (Munn et al., in press), further compromising growth and survival. Therefore, it is easy to appreciate that juvenile red kangaroos have the highest drought-related mortalities of any cohort (Newsome et al., 1967; Bayliss, 1985a; Robertson, 1986; Dawson, 1995). Kirkpatrick and McEvoy found similar age-structured mortality in drought-affected eastern grey kangaroos (Kirkpatrick and McEvoy, 1966), and Arnold et al. suggested that high rates of juvenile mortality are a general feature regulating kangaroo populations (Arnold et al., 1991). Our results provide the first mechanistic explanation linking the physiological constraints faced by juvenile red kangaroos in relation to their drought-related mortalities, rainfall and forage quality, the three principal factors affecting recruitment and overall population dynamics (Bayliss, 1987; Shepherd, 1987; Cairns and Grigg, 1993; Dawson, 1995).

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