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Replacing wheat with canola meal in a partial mixed ration increases the milk production of cows grazing at a restricted pasture allowance in spring

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Abstract. Milk production responses were measured in grazing cows offered supplements in different ways. Holstein–Friesian cows averaging 70 days in milk were allocated into 20 groups of eight, each including one rumen-fistulated cow. One of three dietary treatments was then randomly assigned to each of the 20 groups. Treatments were (1) Control (8 groups), where cows were supplemented with rolled wheat grain fed twice daily in the dairy and pasture silage provided in the paddock; (2) partial mixed ration (PMR; 8 groups), where cows were offered a PMR comprising rolled wheat grain, maize grain, maize silage and lucerne hay, which was presented on a feedpad immediately after each milking; the PMR was formulated to provide the same estimated metabolisable energy intake as the Control supplements; and (3) PMR+Canola (4 groups), where cows were fed in the same way as the PMR cows, except that a proportion of the wheat in the PMR was replaced with solvent-extracted canola meal. This ration was formulated to provide the same metabolisable energy as the Control and PMR treatments, but had greater amounts of crude protein. For Control and PMR treatments, supplements were offered at 8, 10, 12 or 14 kg DM/cow.day (2 groups per amount) while for the PMR+Canola treatment supplement was offered at 12 or 14 kg DM/cow.day (2 groups per amount). In addition to their supplements, all groups grazed an allowance of ~14 kg DM/cow.day (measured to ground level) of perennial ryegrass pasture. Yields of energy-corrected milk increased linearly with increasing supplement intake, but there was no difference between Control and PMR cows. When canola meal was added to the PMR, there was an increase in energy-corrected milk at a predicted supplement intake of 13.0 kg DM/cow.day. This was associated with a greater concentration and yield of milk fat in the PMR+Canola cows. Ruminal fluid pH and DM intake from pasture were also greater in PMR+Canola cows. It is concluded that farmers feeding high amounts of supplements to grazing cows could increase milk production by carefully considering the composition and form of the supplement mix, including the inclusion of canola meal.

Additional keywords: dairy, protein, supplements.

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Introduction

Grazed pasture is an important source of nutrients for dairy cattle in many parts of the world, including south-eastern Australia, because of its inherent low cost (Doyle and Stockdale 2011). To increase per-cow and per-hectare production, pasture is commonly supplemented with grain or pelleted concentrates fed in the dairy, along with conserved forage fed in the paddock (Doyle *et al*. 2000). Below-average rainfall, reduced availability of irrigation water and reduced pasture DM production in recent years in south-eastern Australia (Dairy Australia 2011) have led to increased reliance on bought-in forage and concentrates to meet the nutritional requirements of the milking herd.

Milk production increases in response to cereal and concentrates fed in the dairy at milking times (Walker *et al*. 2001; Leddin *et al*. 2009), but the immediate response is curvilinear, with poorer responses being observed as the amount of grain increases (Stockdale *et al.* 1987; Walker *et al.* 2001; Kellaway and Harrington 2004). Recently, Auldist *et al*. (2013) reported that when the amount of total supplement offered to cows in late lactation in autumn was more than 10 kg DM/cow.day, the immediate milk-production responses were greater when the supplements were offered as a maize-based partial mixed ration (PMR) on a feedpad, than when feeding the same amount of metabolisable energy (ME) as barley grain in the dairy and forage in the paddock. These authors attributed the improved milk response to a more slowly digestible starch source, less variable ruminal fluid pH, more stable and efficient rumen fermentation and increased DM intake (DMI) from pasture in the cows offered PMR.

The current experiment was conducted to determine whether similar improvements in short-term milk production responses to supplements fed as a PMR compared to feeding grain and forage separately could be achieved in early lactation when cows were grazing highly digestible spring pasture. Further, the effects on milk production of replacing some of the wheat in the ration with canola meal were examined. This was undertaken because previous research indicated that protein supplements can be associated with increased DMI and/or milk yield (Gordon 1977; Butler 1998; Broderick 2003; Ipharraguerre and Clark 2005; Olmos Colmenero and Broderick 2006), which was one mechanism contributing to the greater milk production response observed by Auldist *et al*. (2013) in cows offered high amounts of PMR.

The hypotheses tested were (1) that increasing the amounts of grain fed in the milking parlour and forage supplement fed in the paddock to cows grazing a low allowance of pasture in spring would lead to a non-linear (diminishing returns) increase in production of energy-corrected milk (ECM), (2) that increasing the amounts of a PMR formulated to be isoenergetic, but containing a more slowly digestible starch source, will lead to a linear increase in ECM production, and ECM production would be greater than when the supplements were fed separately in the dairy and paddock and (3) that replacing some of the wheat in the PMR with canola meal would further increase DMI and ECM yield. These hypotheses relate to the immediate milk production response and do not consider longer-term 'carry over' benefits from feeding supplements (Broster and Broster 1984).

Materials and methods

The experiment was conducted at the Department of Environment and Primary Industries (DEPI), Ellinbank Centre, Victoria, Australia (38°14′S, 145°56′E). All procedures were conducted in accordance with the Australian Code of Practice for the Care and Use of Animals for Scientific Purposes (National Health and Medical Research Council 2004). Approval to proceed was obtained from the DEPI Agricultural Research and Extension Animal Ethics Committee.

Cows

The experiment was conducted in spring, and used 160 multiparous seasonally calving Holstein–Friesian dairy cows, including 20 rumen-fistulated cows. All cows were between 3 and 7 years old, were 70 ± 15.2 days in milk (mean ± s.d.), and were milked twice daily at ~0700 hours and 1500 hours.

The experiment was conducted over 28 days. This included a 14-day pre-experimental period during which cows adapted to the amount of supplement. After the pre-experimental period, there was a 14-day measurement period during which DMI and milk production were measured.

Dietary treatments

Prior to the experiment, cows were allocated into 20 groups of eight cows, with groups balanced for days in milk, age, liveweight and current production of milk, milk protein and milk fat, according to the method of Baird (1994). Each group included one rumen-fistulated cow. Ten treatments were allocated at random to these 20 groups, two groups per treatment; each group of eight cows was an experimental unit. The 10 treatments consisted of an incomplete factorial of three dietary treatments by four rates of supplementary feed (Table 1). The three dietary treatments were as follows:

(1) Control: cows grazed perennial ryegrass (*Lolium perenne* L.) pasture supplemented with rolled wheat grain fed twice daily in the dairy and pasture silage provided in the paddock. The ratio of grain : forage fed as supplement was 72 : 28 (DM basis). The pasture allowance was ~14 kg DM/cow.day. This treatment was assigned to eight groups.

(2) PMR: cows grazed perennial ryegrass pasture twice daily at the same allowance as the Control cows, and were offered a PMR comprising rolled wheat grain, maize grain, maize silage and lucerne hay (Table 2). The PMR was mixed and chopped in a feed wagon (Richard Keenan and Co. Ltd, Model K160, Co. Carlow, Ireland) before being presented on a concrete feedpad immediately after each milking (half the daily ration was fed following each milking). Water was added to the ration such that the final DM concentration of the ration approximated 50%. The PMR was formulated to provide the same estimated ME intake as the supplements offered to the Control cows, and had approximately the same ratio of grain : forage. This treatment was assigned to eight groups.

(3) PMR+Canola: cows were fed in the same way as the PMR cows, except that a proportion of the wheat in the PMR was replaced with solvent-extracted canola meal (Table 2). This ration was formulated to provide the same supplement ME as the Control and PMR treatments, but had greater amounts of crude protein. This treatment was assigned to four groups.

Establishment of cow requirements and adequacy of diets was based on the CPM Dairy nutrition model (Tedeschi *et al*.

Table 1. Diagrammatic representation of the experimental design, showing grouping of 160 cows into different rates of supplement offered (either 8, 10, 12 or 14 kg DM total supplement/cow.day) for each of three replicated feeding strategies for supplements (Control, partial mixed ration (PMR) and PMR+Canola)

Feeding strategy	Control								PMR								PMR+Canola			
Number of cows	64								64								32			
Replicate number	A				B				A				B				A		B	
Number of cows	32				32				32				32				16		16	
Amount of supplement (kg DM supplement/cow.day)	8	10	12	14	8	10	12	14	8	10	12	14	8	10	12	14	12	14	12	14
Number of cows	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8

Table 2. Composition of Control, partial mixed ration (PMR) and PMR+Canola diets (% of total supplement; DM basis)
Data are means for the 14-day measurement period

Component	Control	PMR	PMR+Canola
Wheat grain	72	39	23
Pasture silage	28	–	–
Maize grain	–	20	20
Maize silage	–	32	32
Lucerne hay	–	9	9
Canola meal	–	–	16

2008). Concentrations of crude protein, acid detergent fibre, neutral detergent fibre, lignin, non-fibre carbohydrate, starch, crude fat, ash and estimated ME in all feed components and pasture (offered, residual and consumed) are presented in Table 3.

Amounts of supplement

Within each dietary treatment, groups were assigned to different amounts of supplement (Table 1). For the Control and PMR cows (of which there were 8 groups of 8 cows), two groups each were assigned to receive 8, 10, 12 or 14 kg DM total supplement/cow.day. For the PMR+Canola cows (of which there were 4 groups of 8 cows), two groups were assigned to receive either 12 or 14 kg DM total supplement/cow.day. Cows on the PMR and PMR+Canola treatments received their supplements on a feedpad, with each of the 12 groups of eight cows separated from the other groups with electric tapes. Individual cows on the Control diet were hand-fed their wheat grain in the dairy at each milking, while the pasture silage was fed by placing the allocation for each group of eight cows under an electric wire in their grazing area each day. Cows receiving the highest amounts of supplement (12 kg and 14 kg DM/cow.day) were introduced gradually to dietary regimens, reaching their full amount of ration by Day 5 of the pre-experimental period.

As part of their supplement, all cows received a vitamin and mineral pellet (Nutrifeed Hi-Milker, Debenham Australia Pty Ltd, Leongatha, Vic., Australia) that contained tylosin (110 mg/100 g pellets) and monensin (110 mg/100 g pellets). Cows at the highest rate of supplementation (14 kg DM/cow.day) received this supplement at the rate recommended by the manufacturers (125 g pellet/cow.day), while cows receiving lower amounts of supplement received proportionally less (e.g. cows offered 8 kg DM supplement/day received 71 g/cow.day of the vitamin and mineral pellet). Control cows received their vitamin and mineral pellets mixed with their grain at milking time, while cows offered PMR and PMR+Canola received their pellets mixed into their PMR.

All cows had several opportunities each day to access water *ad libitum*, from troughs located in and adjacent to the dairy, and in laneways adjacent to the paddocks used for grazing, but they had no access to water while grazing.

Grazing

Pasture allowance was ~14 kg DM/cow.day (to ground level) and was available as two equal allocations of pasture per day (a fresh break after each milking). Control cows had access to pasture immediately after each milking. Cows on the PMR and PMR+Canola treatments were given access to pasture after they had consumed their ration on the feedpad following each milking. Cows grazed in groups of eight on adjacent areas, separated from the other groups by electric tapes. Cows were prevented from re-grazing areas that had been grazed on previous days, by the use of back fencing.

Pasture intake and nutritive characteristics

Pre- and post-grazing pasture mass was estimated every day for each group of eight cows by using a C-Dax pasture meter (Pasturemeter XP1, C-Dax Ltd, Palmerston North, New Zealand). This information was used to calculate average pasture DMI for each group. The C-Dax pasture meter was calibrated for each new set of paddocks the cows entered.

For each new paddock, representative samples of pasture on offer were collected pre-grazing for assessment of pasture nutritive characteristics. Samples of pasture were collected post-grazing from each group of eight cows. All pre- and post-grazing samples were collected by cutting pasture to ground level by using electric shears at several points along a transect of a grazing area.

Pasture samples were thoroughly mixed, then subsampled, washed, freeze-dried and ground through a 0.5-mm sieve. Dried samples were analysed for nutritive characteristics as described for supplement. Data from pre- and post-grazing pasture samples, together with estimates of pre- and post-grazing mass, were used to calculate the nutritive characteristics of the pasture consumed.

Table 3. Mean nutritive characteristics of feed components and pasture
Data are means (% of DM unless otherwise indicated) for all samples collected during the 14-day measurement period. ADF, acid detergent fibre; CF, crude fat; CP, crude protein; ME, estimated metabolisable energy (MJ/kg DM); NFC, non-fibre carbohydrate; NDF, neutral detergent fibre

Feed component	CP	ADF	NDF	Lignin	NFC	Starch	CF	Ash	ME
Crushed wheat grain	16.4	5.5	14.0	1.4	67.0	59.2	2.1	2.7	14.3
Crushed maize grain	9.6	4.2	10.5	1.5	74.6	65.8	4.7	1.7	14.3
Lucerne hay	21.7	34.5	44.9	6.4	26.1	1.2	2.1	9.9	9.7
Pasture silage	16.7	37.3	57.5	5.5	12.6	2.5	5.3	12.2	9.3
Maize silage	8.8	23.5	39.4	3.3	43.6	34.8	3.6	5.8	10.7
Canola meal	42.0	18.5	31.7	8.2	22.9	2.9	2.9	7.0	12.5
Pasture offered	14.6	31.5	57.0	3.2	21.6	3.0	3.1	7.7	10.3
Pasture residual	11.2	38.7	64.6	5.3	17.3	1.7	2.1	8.9	8.5
Pasture consumed	17.0	26.4	51.5	2.1	24.6	4.0	3.8	7.0	11.5

Supplement intake and nutritive characteristics

Samples of the ration components (wheat grain, maize grain, maize silage, lucerne hay, canola meal and pasture silage) were collected on 3 days/week during the measurement period and bulked by week. Each sample was frozen, freeze-dried, ground through a 0.5-mm sieve and analysed at a commercial laboratory (Dairy One Forage Laboratory, Ithaca, NY, USA) for nutritive characteristics by near-infrared spectroscopy (Method 989.03; AOAC International 2000). Concentrations of estimated ME were calculated using the following formula (National Research Council 2001):

$$\mathrm{ME\,(MJ/kg\,DM)} = (((1.01 \times (0.04409 \times \mathrm{TDN})) - 0.45) \times 4.184,$$

where TDN is total digestible nutrient (%).

Wheat grain (Control cows) and ration (PMR and PMR+Canola cows), offered and refused, were weighed every day of the measurement period. Samples of grain and ration refused by each group of cows were collected every day of the measurement period and analysed for DM and nutritive characteristics. This allowed the calculation of daily DMI, and intake of estimated ME, crude protein and neutral detergent fibre for each group of cows. No refusals of pasture silage, fed to groups of cows in the plots (Control cows), were observed.

Milk yield and composition

Milk yield of every cow was measured at every milking by using a DeLaval Alpro milk metering system (DeLaval International, Tumba, Sweden). A composite sample of the daily milk (evening + morning) was taken on two occasions per week during the measurement period, using in-line milk meters (DeLaval International). Milk samples were tested for concentrations of protein and fat by using an infrared milk analyser (Model 2000, Bentley Instruments, Chaska, MN, USA). ECM was calculated using the following formula (Tyrrell and Reid 1965):

$$\mathrm{ECM\,(kg/cow.day)} = \mathrm{milk\ yield\ kg} \times (376 \times \mathrm{fat\%} + 209 \times \mathrm{protein\%} + 948)/3138.$$

Body condition score and liveweight

Body condition score (BCS) was assessed on two consecutive days immediately before and after the experiment. This was performed by two trained assessors according to the 8-point scale of Earle (1976). Cows were weighed at the same time.

Ruminal-fluid pH and concentrations of volatile fatty acids (VFA) and ammonia

Samples of rumen fluid were collected from each of the rumen-fistulated cows at intervals of ~2 h over a 24-h period during the measurement period. This was performed by restraining the cows in temporary yards set up in the paddock, or in permanent yards next to the feedpad and dairy. Samples were collected *per fistula* by using a 100-mL plastic syringe connected to a copper pipe inserted into the rumen. Fluid was collected from several sites within the rumen. Samples were immediately analysed for pH by using a portable pH meter (Mettlet-Toledo FG2 pH meter, Schwerzenbach, Switzerland).

For VFA analyses, an aliquot of 4 mL of ruminal fluid was dispensed into a tube containing 1 mL of 25% metaphosphoric acid, before being stored at −20°C until subsequent analysis. Concentrations of VFA were determined by capillary gas chromatography using the method of Packer *et al.* (2011). Sample VFA peaks were identified by comparing their retention time with those of a standard mixture of VFA (Sigma Aldrich Pty Ltd, Castle Hill, NSW, Australia) and quantified using Shimadzu Class GC10 Version 1.62 (Shimadzu Scientific Instruments, Rydalmere, NSW, Australia), with 4-methylvaleric acid as the internal standard. Results were calculated as ppm and converted to mmol/L. The ratio of acetate plus butyrate to propionate ((A+B) : P) was calculated from molar concentrations.

For ammonia-N analysis, an aliquot of 10 mL of ruminal fluid was dispensed into a tube and stored at −20°C until analysis. Concentrations of ammonia were assayed by a direct enzymatic procedure using a commercially available kit (Boehringer Mannheim, R-Biopharm Laboratory Diagnostics Pty Ltd, Taren Point, NSW, Australia) and a Cobas Mira S autoanalyser (Roche, Montclair, NJ, USA).

Statistical analyses

Statistical analyses were conducted on treatment-period data for each group of eight cows (the experimental unit). Intake, liveweight and BCS data (group averages) were analysed by ANOVA in Genstat for Windows (Genstat release 14, VSN International, Hemel Hempstead, UK). The treatment structure for the ANOVA was factorial, treatment by supplement rate, nested within a binary factor indicating the presence/absence of canola, with group as the experimental unit. A contrast was defined within the ANOVA to compare PMR+Canola with PMR at supplement rates 12 and 14 kg. Hence, hypothesis *P*-values were available for of (1) the main effect of PMR (excluding PMR+Canola) vs Control, (2) the main effect of PMR+Canola vs PMR, (3) the main effect of supplement rate, controlling for dietary treatment, and (4) the interaction between dietary treatment and supplement rate.

Milk yield, ECM yield, fat concentration and yield and protein concentration and yield group means were derived before statistical analysis by averaging over the treatment period within cows and averaging these across cows within groups. The group mean data were then analysed as responses to observed DMI by using REML in Genstat for Windows (Genstat release 14, VSN International, Hemel Hempstead, UK). The fixed effects in the REML model consisted of the factor treatment (Control, PMR or PMR+Canola) by a second-order orthogonal polynomial for DMI:

$$y = \mu + \alpha_i + \beta_i x_1 + y_i x_2 + \varepsilon,$$

where y is the response variable, α, β and γ are constant, linear and quadratic coefficients, respectively, that depend on Dietary treatment i = Control, PMR, PMR+Canola, (with $\gamma \equiv 0$ for PMR+Canola), x_1 and x_2 are linear and quadratic orthogonal polynomial transforms of DMI, and ε is a random group effect. The purpose of the linear and quadratic orthogonal polynomial terms was to enable a clear distinction to be drawn between linear trend and curvature of responses to DMI. Cumulative *F*-tests were used to test the significance of dietary treatment, linear and quadratic relationships with DMI, and linear and quadratic interactions between DMI and dietary treatment.

Table 4. Pre-grazing and residual pasture mass (t DM/ha), pasture allowance (kg DM/cow.day), daily DM intake (DMI; kg DM/cow.day) and estimated intake of metabolisable energy (ME; MJ/cow.day) from pasture and supplement for cows offered the Control, partial mixed ration (PMR) and PMR +Canola supplements at nominal amounts of 8, 10, 12 or 14 kg DM/cow.day

P (treat), *P*-value comparing PMR (excluding PMR+Canola) with Control, averaged over the four supplement rates. *P* (rate), *P*-value for the main effect of rate of supplement feed offered, controlling for feeding strategy. *P* (canola), *P*-value comparing PMR+Canola with PMR, restricted to supplement rates 12 and 14 kg DM/cow. Data are means from the 14-day measurement period

Treatment	Supplement offered	Pre-grazing pasture mass	Post-grazing pasture mass	Pasture allowance	Pasture		Supplement		Total intake			
					DMI	ME	DMI	ME	DMI	ME	NDF	CP
Control	8	4.61	1.42	14.4	10.0	110	7.7	100	17.7	211	7.07	2.90
	10	4.39	1.59	13.7	8.7	102	9.6	126	18.4	228	6.87	3.08
	12	4.59	1.99	14.3	8.1	94	11.6	152	19.7	245	7.07	3.31
	14	4.49	2.12	14.0	7.4	86	13.4	176	20.8	261	7.08	3.49
PMR	8	4.38	1.64	13.7	8.6	96	8.8	114	17.4	209	6.67	2.59
	10	4.61	1.83	14.4	8.7	98	10.8	140	19.5	238	7.30	2.87
	12	4.56	2.15	14.2	7.5	89	12.9	167	20.4	256	6.89	3.02
	14	4.17	2.18	13.0	6.2	76	14.8	192	21.0	268	6.49	3.09
PMR+Canola	12	4.55	1.88	14.2	8.4	99	13.0	170	21.4	268	7.16	3.67
	14	4.42	1.91	13.8	7.8	91	15.1	197	22.9	288	7.72	3.85
P (treat)		0.368	0.048	0.368	0.036	0.052	<0.001	<0.001	0.236	0.134	0.477	<0.001
P (rate)		0.419	<0.001	0.419	0.003	0.012	<0.001	<0.001	<0.001	<0.001	0.732	<0.001
P (canola)		0.361	0.029	0.361	0.024	0.041	0.033	0.007	0.013	0.016	0.063	<0.001
s.e.d.		0.19	0.15	0.58	0.66	7.50	0.12	1.57	0.68	7.93	0.505	0.107

(A special factor for the two levels of supplementary feeding within PMR+Canola was included first to ensure that the tests of significance for dietary strategy, rate, and their interaction, related to the Control and PMR treatments only.) Fitted curves were re-expressed and reported as simple quadratic or linear functions of DMI. Random effects in the REML model consisted simply of group.

Data for VFA (for which there was one fistulated cow per group) were analysed by ANOVA with factorial treatment structure, treatment by supplement rate by interval, and nested blocking structure, herd split for cow, split for interval.

Time was measured from the beginning of morning milking, and rumen pH data were linearly interpolated over intervals (on average, length 2 h) between measurement times for a 24-h period. Using the interpolated data, the following were calculated for each (fistulated) cow: time under pH 6, area under pH 6 (pH × h), maximum pH, minimum pH, mean pH, and minimum pH within the first 7 h (pH nadir). These summary statistics were subjected to ANOVA, as per the intake data.

Residuals were checked graphically for normality of distribution and constant variance for each analysis.

Results

Pasture allowances and intakes

By design, neither pre-grazing pasture mass nor pasture allowance per cow differed among diets or amount of supplement offered (Table 4). Post-grazing pasture mass generally increased as the amount of supplement offered increased, and was greater for PMR cows than Control cows but lower for PMR+Canola cows than for PMR cows. As a consequence, estimated pasture DM and ME intake decreased as the amount of supplement offered increased. Pasture DMI was greater for Control cows than for PMR cows, but there was no difference in intake of ME from pasture. Pasture DMI and intake of ME from pasture were greater for PMR+Canola cows than for PMR cows.

Supplement intakes and total intakes

Both estimated supplement DM and ME intake increased as the amount of supplement offered increased. They were also affected by diet treatment, being lower for Control cows than for PMR cows (Table 4). Supplement DMI was greater for PMR+Canola cows than for PMR cows, but ME intake from supplement was not. Total DMI and apparent ME intakes were not different among diets, but increased with increasing amounts of supplement offered.

Milk yield and composition

Mean yields of milk, ECM, fat and protein, and mean concentrations of milk fat and protein, for cows on the three dietary treatments at the different supplement levels are presented in Fig. 1. Fitted curves are also presented for each of these variables for the Control and PMR treatments; the equations describing them are given in Table 5. There was no difference ($P > 0.05$) between PMR and Control cows for milk yield or composition at any amount of supplement intake. However, there were differences between the PMR+Canola cows and the other two dietary treatments. Specifically, at a predicted supplement intake of 13.0 kg DM/cow.day, fat concentration, fat yield and ECM yield was greater ($P < 0.05$) for PMR+Canola cows than for Control and PMR cows (Fig. 1). At a predicted supplement intake of 15.1 kg DM/cow.day, fat yield was greater ($P < 0.05$) for PMR+Canola cows than for Control cows.

BCS and liveweight

At the end of the experiment, there were no differences among the dietary treatments in BCS or liveweight, nor in the change in BCS or liveweight during the experiment (Table 6). Cows had greater

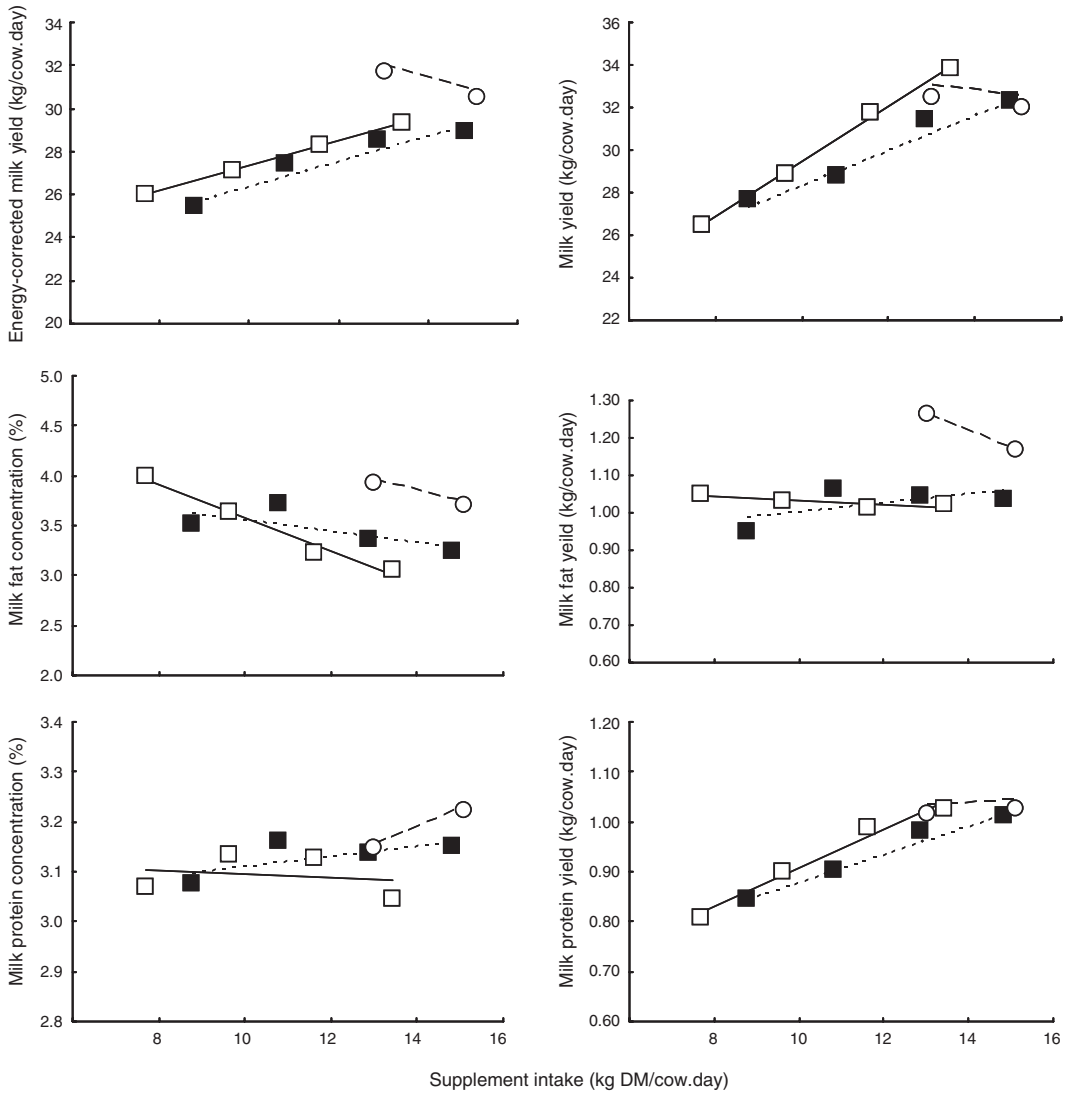

Fig. 1. Mean daily yields of milk and energy-corrected milk (ECM), and concentrations and yields of milk protein and fat, for cows offered either the Control (□), partial mixed ration (PMR, ■) or PMR+Canola (○) diets at amounts of ~8, 10, 12 or 14 kg DM supplement/cow.day. Data are means from the 14-day measurement period. Curves were fitted for the Control (solid line) and PMR (short-dashed line) diets.

gain in BCS and greater final BCS as the intake of supplement increased. There was a trend for cows to gain more liveweight as the amount of supplement increased.

Ruminal-fluid pH, VFA and ammonia

Features of the daily variation in ruminal pH for cows offered the three diets are presented in Table 7. There were no differences between the Control and PMR diets for time under pH 6.0, area under pH 6.0, minimum pH, or mean daily pH. Maximum pH was greater for PMR cows than for Control cows. Area under pH 6.0 increased as the amount of supplement increased, but minimum pH decreased. Adding canola to the PMR decreased area under pH 6.0 and increased minimum pH in those cows compared to PMR cows. Concentrations of total VFA for cows, along with proportions of acetate, propionate, butyrate and valerate, ammonia and the (A+B)/P ratio in ruminal fluid from cows on the 3 diets at the different levels of intake are presented in Table 8. Dietary treatment had no effect on any of these variables. The proportions of butyrate, the (A+B)/P ratio and the concentration of ammonia in ruminal fluid decreased as the amount of supplement offered increased. Proportions of butyrate, the (A+B)/P ratio and concentrations of ammonia were greater in PMR+Canola in cows compared to PMR cows, but proportions of propionate were lower.

Discussion

The first hypothesis tested in the present experiment was that increasing the amount of supplement fed as grain in the dairy and forage in the paddock to cows grazing a low allowance of pasture would lead to a non-linear increase in the production of

Table 5. Equations describing the relationships between supplement DMI (kg DM total supplement/cow.day) and milk yield (MY), energy-corrected milk yield (ECM), fat concentration (F%), fat yield (FY), protein concentration (P%) and protein yield (PY), for cows offered the Control and partial mixed ration (PMR) supplements

These equations were derived from curves fitted to the mean measured values at four levels of supplement intake per diet. Quadratic contrasts were not significant and are not shown)

Parameter	Diet	Equation	*P*-value of linear trend
MY (kg/cow per day)	Control	MY = 16.59 + 1.29DMI	<0.001
	PMR	MY = 20.17 + 0.81DMI	<0.001
ECM (kg/cow per day)	Control	ECM = 22.87 + 0.44DMI	<0.001
	PMR	ECM = 21.24 + 0.51DMI	<0.001
F%	Control	F% = 5.254 – 0.168DMI	0.001
	PMR	F% = 4.115 – 0.057DMI	<0.001
FY (kg/cow per day)	Control	FY = 1.084 – 0.005DMI	<0.001
	PMR	FY = 0.881 + 0.012DMI	<0.001
P%	Control	P% = 3.134 – 0.004DMI	0.728
	PMR	P% = 3.002 + 0.011DMI	0.322
PY (kg/cow per day)	Control	PY = 0.523 + 0.039DMI	<0.001
	PMR	PY = 0.592 + 0.028DMI	<0.001

ECM. This was based on previous research indicating that marginal responses to supplementary cereal-grain decline with increasing amounts of grain fed, and become negative when between 5 and 10 kg DM of cereal grain are fed (Walker *et al*. 2001; Beever and Doyle 2007). In the current experiment, this was not observed; the production of milk and ECM by the Control cows increased linearly to an intake of 14 kg DM of total supplement. Thus, the first hypothesis is not supported. A supplement intake of 14 kg DM total supplement/cow per day (10.5 kg DM of wheat grain) is substantially greater than would be typically fed on dairy farms in south-eastern Australia. Why such a high amount of grain did not elicit a reduction in milk response is uncertain, since results of the rumen-fluid analyses would suggest that a decline could have been expected. However, it is noted that the cows had been acclimatised to high amounts of grain from calving until the start of the experiment (an average of ~10 weeks).

Feeding cows supplementary grain and forage as a PMR that was isoenergetic with the Control diet also resulted in a linear increase in ECM production as total supplement intake increased from 8 to 14 kg DM/cow.day. Thus, the first part of the second hypothesis was supported. However, there were no differences in milk production between the two diets at any level of intake, thus negating the second part of the second hypothesis. Auldist *et al*. (2013) reported an increased marginal milk production response to a maize-based PMR, compared with a barley-based Control diet in cows grazing at a restricted pasture allowance in autumn, when cows were in late lactation. In that experiment, however, the Control cows had maximum ECM production at 10.0 kg DM total supplement/cow.day, and the marginal ECM response declined after that. Thus, a major factor in the lack of difference in milk production between the Control and PMR diets in the current experiment was that intake was not high enough to compromise rumen function in the Control cows, or for the advantages of the PMR to be manifested as increased milk production. This was

Table 6. Mean body condition score (BCS) and liveweight (LW) before and after the experiment for cows offered the Control, partial mixed ration (PMR) and PMR+Canola supplements at nominal amounts of 8, 10, 12 or 14 kg DM/cow.day

P (treat), *P*-value comparing PMR (excluding PMR+Canola) with Control, averaged over the four supplement rates. *P* (rate), *P*-value for the main effect of rate of supplement feed offered, controlling for feeding strategy. *P* (canola), *P*-value comparing PMR+Canola with PMR, restricted to supplement rates 12 and 14 kg DM/cow

Parameter	Control				PMR				PMR+Canola		*P* (treat)	*P* (rate)	*P* (canola)	s.e.d.
	8	10	12	14	8	10	12	14	12	14				
BCS pre-experiment	4.42	4.49	4.41	4.41	4.39	4.39	4.47	4.38	4.39	4.45	0.470	0.627	0.931	0.062
BCS post-experiment	4.36	4.57	4.44	4.54	4.23	4.39	4.59	4.51	4.42	4.60	0.236	0.005	0.533	0.077
BCS change	–0.06	0.08	0.03	0.13	–0.16	0.00	0.12	0.13	0.03	0.15	0.602	0.022	0.619	0.086
LW pre-experiment	542	561	548	561	534	538	562	534	546	563	0.178	0.491	0.553	15.11
LW post-experiment	548	569	571	583	538	548	574	561	569	581	0.135	0.107	0.511	15.71
LW change	6	8	24	20	3	10	12	27	22	19	0.702	0.062	0.868	8.45

Table 7. Features of the daily variation in the pH of ruminal fluid for cows offered the Control, partial mixed ration (PMR) and PMR+Canola supplements at nominal amounts of 8, 10, 12 or 14 kg DM/cow.day

P (treat), *P*-value comparing PMR (excluding PMR+Canola) with Control, averaged over the four supplement rates. *P* (rate), *P*-value for the main effect of rate of supplement feed offered, controlling for feeding strategy. *P* (canola), *P*-value comparing PMR+Canola with PMR, restricted to supplement rates 12 and 14 kg DM/cow. Data are means of two rumen-fistulated cows per rate of supplement per diet

Parameter	Control				PMR				PMR+Canola		*P* (treat)	*P* (rate)	*P* (canola)	s.e.d.
	8	10	12	14	8	10	12	14	12	14				
Time under pH 6.0 (days)[A]	0.38	0.48	0.44	0.44	0.30	0.44	0.45	0.47	0.34	0.39	0.466	0.059	0.066	0.062
Area under pH 6.0 (pH × h)[B]	2.77	2.81	2.44	4.05	1.43	2.82	4.79	4.11	2.00	2.07	0.502	0.022	0.001	0.771
Maximum pH	6.73	6.74	6.83	6.95	7.11	7.00	7.04	7.06	7.03	7.15	0.008	0.539	0.702	0.143
Minimum pH	5.52	5.53	5.43	5.24	5.45	5.55	5.31	5.27	5.62	5.55	0.565	0.045	0.009	0.130
Mean daily pH	6.13	6.10	6.15	6.16	6.26	6.09	6.12	6.15	6.22	6.21	0.432	0.120	0.057	0.054

[A]Mean time per day during which ruminal pH was below 6.0.
[B]Area of the pH vs time of day curve below pH 6.0 (pH × h).

Table 8. Mean daily volatile fatty acid (VFA) concentrations in ruminal fluid of cows offered the Control, partial mixed ration (PMR) and PMR+Canola supplements at nominal amounts of 8, 10, 12 or 14 kg DM/cow.day

P (treat), *P*-value comparing PMR (excluding PMR+Canola) with Control, averaged over the four supplement rates. *P* (rate), *P*-value for the main effect of rate of supplement feed offered, controlling for feeding strategy. *P* (canola), *P*-value comparing PMR+Canola with PMR, restricted to supplement rates 12 and 14 kg DM/cow. Data are means of two rumen-fistulated cows per rate of supplement per diet

Parameter	Control				PMR				PMR+Canola		*P* (treat)	*P* (rate)	*P* (canola)	s.e.d.
	8	10	12	14	8	10	12	14	12	14				
Total VFA (mmol/L)	129.7	125.6	121.1	128.7	111.7	128.3	118.0	123.1	122.2	118.5	0.067	0.401	0.954	5.84
Acetate (%)	65.2	62.2	60.6	58.9	63.7	64.0	60.9	59.9	65.7	63.5	0.764	0.093	0.059	2.80
Propionate (%)	17.4	23.6	25.8	27.8	18.8	19.3	26.6	25.3	17.5	18.2	0.613	0.060	0.025	4.37
Butyrate (%)	14.1	10.5	9.1	7.9	12.7	11.5	8.2	10.2	13.4	14.1	0.787	0.028	0.010	2.02
Valerate (%)	1.5	1.5	1.4	0.8	2.4	2.0	1.2	1.5	1.5	2.0	0.119	0.225	0.327	0.57
(A+B)/P[A]	4.6	3.4	2.8	2.5	4.2	4.0	2.8	2.9	4.6	4.3	0.690	0.028	0.012	0.75
Ammonia (mg/L)	194	115	107	77	104	125	58	64	168	142	0.078	0.039	0.004	35.9

[A]Molar proportions of acetate (A), butyrate (B) and propionate (P).

supported by the ruminal-fluid pH data that showed no difference among diets in terms or daily pH, minimum pH nor time spent below pH 6.0. Further, although concentrations of milk fat declined with increasing supplement intake for all groups, there was no difference between the Control cows and PMR cows at any level of intake. This is in contrast to the experiment of Auldist *et al*. (2013) where feeding supplement as a PMR arrested the decline in milk fat seen in the Control cows as supplement intake increased. These authors speculated that this effect was due to numerically lower ruminal-fluid pH in the Control cows causing a shift in the rumen microbial population and altered rumen lipid metabolism (Bauman and Griinari 2003). This is further evidence that such differences in the rumen environment of Control and PMR cows were not present in the current experiment. Moreover, the lack of difference in milk production is mirrored by the lack of effect of diet on BCS and liveweight, even though 28 days is a short time frame in which to measure changes in these parameters.

When some of the wheat in the PMR was replaced by canola meal, yields of ECM were increased, thereby supporting the third hypothesis. Presumably, this effect was driven at least in part by the increased crude protein of the PMR+Canola ration (total diet crude protein 16.2%) compared with the PMR ration (total diet crude protein 14.1%), as increasing crude protein intake through the provision of canola and other protein supplements has increased milk yield in previous experiments (Oldham 1984; Huhtanen *et al*. 2011; Martineau *et al*. 2013). Part of the increased ECM yield of PMR+Canola cows in the current experiment was due to an increased pasture intake of these cows compared with PMR cows. Protein supplements such as soybean meal and canola meal have previously been associated with increased DMI (Butler 1998; Broderick 2003; Ipharraguerre and Clark 2005; Olmos Colmenero and Broderick 2006) via a variety of mechanisms. One possibility is that high-protein feeds have greater buffering capacity in the rumen than do low-protein forages (Allen *et al*. 2006), thus acting to stabilise ruminal-fluid pH to a relatively greater degree and, in turn, leading a greater inclination to eat. There is support for this mode of action in the ruminal-fluid pH data of the current experiment, which showed that adding canola meal to the ration decreased the amount of time per day ruminal-fluid pH spent under pH 6.0 and increased daily minimum pH. This effect occurred in spite of the lack of effect of the canola meal on total VFA production and, in addition to stimulating pasture DMI, may have allowed for faster and more efficient fibre digestion and extraction of energy from the total diet (Mould *et al*. 1983).

Another possible mode of action for the increased ECM yield of PMR+Canola cows compared with PMR cows is that the additional crude protein provided by the canola meal resulted in an increased and more balanced supply of amino acids, which enhanced milk production and, as a result of the increased energy demand, increased DMI ('pull effect'; Huhtanen *et al*. 2011).

Whatever the mechanism, the increased milk yield associated with adding canola meal to the PMR (and thereby increasing the crude protein concentration of the diet) may help explain why there were no differences in milk production between the Control and PMR cows. In the present experiment, the crude protein concentration of the wheat grain offered to the Control cows was high (16.5%), which resulted in the average crude protein of the total diet of the Control cows (16.7%) being greater than for the PMR cows (14.8%). Thus, one reason that feeding supplements to cows as PMR did not show any advantage to feeding grain in the dairy and forage in the paddock could have been the difference in crude protein intake, even though ME intakes were similar.

In conclusion, we did not see any benefit of feeding supplements as a PMR compared with feeding the same amount of dietary energy as grain in the dairy and forage in the paddock, at least up to 14 kg DM total supplement/cow.day for cows in early lactation. However, when canola meal was added to the PMR, which brought the total diet crude protein up to a concentration equivalent to that of the Control diet, an increase in ECM was achieved in the PMR+Canola cows compared with the Control cows. This was due in large part to a greater fat concentration in those cows, which was presumably associated with a more stable ruminal-fluid pH in the PMR+Canola cows. Farmers feeding high amounts of supplements to grazing cows could increase milk production by carefully considering the composition and form of the supplement mix. The profitability of these strategies will obviously depend on the prevailing cost of canola in comparison to other feedstuffs, the additional costs of implementing a PMR system, and the milk price, which warrants further analyses.

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