

Controlling voluntary intake of molasses-based supplements in grazing cattle

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Abstract. Molasses-based liquid supplements fed *ad libitum* are widely used to provide additional metabolisable energy, non-protein N (NPN) and other nutrients to grazing cattle, but it is often difficult to achieve target intakes of supplementary nutrients. Experiments examined the effects of increasing concentrations of phosphoric acid, urea and ammonium sulfate on the voluntary intake (VI) of molasses-based supplements offered *ad libitum* to heifers grazing tropical pastures. In Experiment 1, the VI of a supplement containing 78 g urea/kg and 26 g phosphoric acid/kg as-fed (M80U+PA) was 3.61 g DM/kg liveweight (LW) per day, and provided 181 mg NPN and 32.4 mg phosphorus (P)/kg LW per day. Increasing the urea content of the supplement to 137 g/kg (M140U+PA) or 195 g/kg (M200U+PA) reduced VI of supplement DM, NPN and P by up to 76%, 44% and 80%, respectively. VI of supplement containing ammonium sulfate (M140+AS+PA) was lower ($P < 0.05$) than that of M140U+PA supplement, and tended ($P > 0.05$) to be lower than that of M200U+PA supplement. In experiment 2, the VI by heifers of a supplement containing 200 g urea/kg (M200U) was 1.53 g supplement DM/kg LW per day, which provided 186 mg NPN/kg LW per day. Inclusion of 49 g phosphoric acid/kg as-fed in this supplement (M190U+50PA) reduced ($P < 0.05$) VI of supplement DM and NPN by 33% and 36%, respectively, while inclusion of 97 g phosphoric acid/kg (M180U+100PA) reduced ($P < 0.05$) VI of supplement DM and NPN by 43% and 48%, respectively. The M190U+50PA and M180U+100PA supplements provided 16 and 26 mg P/kg LW per day, respectively. Heifers not fed supplements gained 0.07 kg/day, and the M200U supplement increased ($P < 0.05$) LW gain to 0.18 kg/day. LW gain was further increased ($P < 0.05$) by the M190U+50PA to 0.28 kg/day, indicating a growth response to supplementary P. No adverse effects of the supplements on animal health were observed in any of the experiments. In conclusion, addition of urea and/or phosphoric acid to molasses supplements effectively reduced VI of supplementary DM, NPN and P, and in the circumstances of Experiment 2, both molasses-urea and P supplements increased heifer LW.

Additional keywords: animal growth, phosphoric acid, supplement acidity, supplement intake, urea.

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Introduction

Production of grazing cattle is often constrained by low concentrations of nutrients in the pasture. For example, in the seasonally dry tropics of northern Australia, the Americas and Africa, pastures are often low in digestibility and thus metabolisable energy (ME), deficient in nitrogen (N) and sulfur during the dry season, and deficient in minerals such as phosphorus (P), sulfur, sodium and cobalt during the wet season (Little 1982; McDowell *et al.* 1984; Winks 1984). Supplementation can alleviate such nutritional inadequacies, but it is often difficult to manage delivery systems for grazing animals so that target amounts of supplementary nutrients are ingested. In extensive grazing systems, such supplements are usually provided *ad libitum* as liquid supplements, loose mixes or blocks, and the voluntary intake (VI) of the supplement is constrained by a low palatability of the supplement and/or the physical restrictions to intake of the supplement (McDowell

1996; Bowman and Sowell 1997; Dixon *et al.* 2003). Effective manipulation of the attractiveness and the VI of supplements is clearly essential to achievement of target intakes of supplements by grazing cattle across a wide range of circumstances.

Molasses is widely used as an attractant and a carrier for supplementary nutrients such as non-protein N (NPN) and minerals, and to provide additional ME (Wythes and Ernst 1984; Bowman *et al.* 1995). Although molasses can be fed as solidified feed blocks (Leng 1984), the manufacturing substantially increases the cost. Thus, molasses is often fed as a liquid supplement. Lick wheels, roller drums and floats can be used to constrain VI of the supplement by limiting access and rate of intake such as by utilising tongue-fatigue of the animal to constrain the amounts ingested. An alternative is to include ingredients that reduce the palatability and thus the VI of the supplement. Molasses-based supplements that include phosphoric or hydrochloric acids utilise the sour flavour for

ruminants to constrain VI (Goatcher and Church 1970). These are widely used commercially for grazing cattle in northern America and Australia (Klett *et al.* 2000). Alternatively, the bitter flavour (Goatcher and Church 1970; Grovum and Chapman 1988) and the conditioned flavour aversions that can be induced by urea (Chalupa *et al.* 1979) have been used to constrain the VI of molasses-based supplements, while the urea also provides NPN. The latter approach has been widely used in the extensive cattle industry of north-eastern Australia, where molasses is readily available, to supplement grazing cattle during drought and for increased production. VI of molasses-urea mixtures containing 7–11% urea and provided *ad libitum* in open troughs usually ranges from 4 to 10 g as-fed/kg liveweight (LW) (Wythes and Ernst 1984; Dixon *et al.* 2003). Urea toxicity seldom occurs, providing that some simple management precautions are applied; these include that the urea is completely dissolved in the molasses, that hungry cattle are not allowed access to the supplement, and animals that are not allowed to drink rainwater lying on the supplement. P supplements can also be provided using this delivery system by inclusion of soluble P into the supplement (McCosker and Winks 1994).

Although provision of molasses supplements in open troughs has important advantages, a disadvantage is that VI of supplement DM and NPN is generally much higher than can be achieved using lick-wheel or block-supplement delivery systems. Also, when molasses supplements containing 7–11% urea are offered in open troughs, the VI of NPN as urea is usually considerably in excess of the amount of rumen-degradable N required for rumen fermentation of the molasses and to provide for a protein deficiency of the grazed pasture. Limited information is available to evaluate the effects of inclusion of acids and high concentrations of urea and other NPN sources, alone or in combination, on the VI of molasses-based supplements offered *ad libitum*. VI of molasses-based supplements has been inversely related to urea concentration (Beames 1960; Silvestre *et al.* 1977) and concentrations up to 20% urea have been fed successfully (Ramirez and Sutherland 1971; Dixon and Smith 2000).

Knowledge of the factors controlling the VI of molasses-based supplements is needed to be able to control the VI of molasses-based supplements by grazing cattle and to provide optimal amounts of supplementary nutrients. The present study examined the effects of various concentrations on the VI of molasses-based supplements by grazing cattle, and in one experiment the effects of supplements containing high concentrations of urea and phosphoric acid on LW change were measured.

Materials and methods

General

The experiments were conducted at the Swan's Lagoon Research Station situated 100 km SSE of Townsville in the seasonally dry tropics of northern Australia. Rainfall at this site is summer dominant; the long-term rainfall distribution and that during the experiments are shown in Table 1. The pastures comprised tropical grasses native or naturalised to the open eucalyptus woodlands of the speargrass region of coastal north-eastern Australia. Major species were black speargrass (*Heteropogon contortus*), with other tropical tall and medium

Table 1. Monthly rainfall preceding and during the experiments and the 34-year average for the site

Month	1999/2000	2000/2001	34-year average
July	0	0	15
August	0	0	19
September	0	0	9
October	0	44	30
November	79	139	67
December	207	266	123
January	26	61	195
February	480	86	187
March	60	57	114
April	108	0	44
May	101	21	40
June	40	0	18
Total	1101	674	871

grasses including *Chrysopogon fallax* and *Bothriochloa pertusa*. The seasonal cycles of pasture growth, nutritional quality of pasture and cattle growth have been described by Winks (1984). The amount of pasture on offer at the commencement of each of the experiments was estimated to exceed 3 t DM/ha.

Bos indicus × *Bos taurus* (>F₂) heifers from the research station herd used in the experiments were generally of docile temperament and accustomed to mustering and handling. The heifers had been offered molasses-based supplements for 1–2 weeks while held in yards immediately after weaning ~8 months before the experiments commenced. In each experiment, 60 heifers were allocated by stratified randomisation based on LW to 12 15-ha paddocks. These paddocks were considered as three blocks on the basis of soil type, vegetation and burning history, and the supplementation treatments were allocated randomly to paddocks within blocks. Full LW was measured following an early morning muster and before the heifers had access to water. Fasted LW was measured after the heifers had been held in yards for 24 h with access to water but without feed. Body condition score was estimated on a 9-point scale (NRC 1996).

The supplements were provided in a cylindrical feed trough 0.56 m in diameter located in a 3 by 3 m shed ~50 m from the only waterpoint in each paddock. The amount of supplement remaining was weighed twice weekly, and additional supplement was added as necessary to maintain *ad libitum* availability. The molasses and urea components of the supplements were mixed in a horizontal paddle mixer until all of the urea was dissolved, and the other ingredients were then added. Acidity of supplements was determined following dilution of the sample with an equal volume of distilled water by using a glass electrode pH meter (Piccolo ATC, Hanna Instruments, Woonsocket, RI, USA). DM content of supplement was determined by drying at 100°C. The amount of supplementary NPN ingested in excess of that required to ferment the molasses in the supplement was calculated on the assumption that 14 g of rumen-degradable N was required per kg molasses and the urea N was used with an efficiency of 0.8 (CSIRO 2007). The data were analysed statistically by ANOVA where the paddock group was considered as the experimental unit (GENSTAT 5, Release 4.1, 4th edition, VSN International, Hemel

Hempstead, UK). Planned comparisons among means were made using a protected least significant difference test.

Experiment 1. Addition of various concentrations of urea or ammonium sulfate to molasses supplements

Sixty heifers, initially 12–16 months of age, with LW mean of 209 kg (s.d. 9 kg) and body condition score mean of 5.6 (s.d. 0.4), were allocated to their paddocks and supplementation treatments commenced in late March 2000 in the late wet season. Three supplement treatments consisted of mixtures of molasses, urea and phosphoric acid (in the form of technical grade orthophosphoric acid). Treatments M80U+PA, M140U+PA and M200U+PA contained 78, 137 and 195 g urea, and 24–26 g phosphoric acid/kg as-fed supplement (Table 2). A fourth treatment (M140U+AS+PA) contained the same amount of urea/kg molasses as did the treatment M140U+PA, and also ammonium sulfate (120 g/kg as-fed supplement) and additional water; the total NPN was the same per kg of as-fed as for supplement M200U+PA. To prepare the M140U+AS+PA supplement, it was necessary to dissolve the ammonium sulfate in water (145 g/kg as-fed supplement) for 16 h before addition to the other ingredients. The heifers allocated to the supplement treatment M140U+AS+PA were offered M200U+PA during Week 1 and then M140U+AS+PA from Week 2. Supplements were fed for 8 weeks.

Experiment 2. Supplement intake and liveweight gain of heifers fed no supplement, or molasses-based supplements containing urea without or with phosphoric acid

Sixty heifers, initially 12–16 months of age, with LW mean of 191 kg (s.d. 11 kg) and body condition score mean of 5.6 (s.d. 0.4), were used for this experiment which commenced in late February 2001 in the mid-wet season. The four supplement treatments consisted of no supplement (Nil), molasses–urea supplement containing 200 g urea/kg as-fed supplement (M200U), or this latter supplement including 49 or 97 g phosphoric acid (as technical grade orthophosphoric acid) per kg supplement (M190U+50PA; M180U+100PA) (Table 3). Supplements were continued for 15 weeks until June, into the early dry season. Full LW was measured at the beginning, after 6 weeks and at the end of the experiment, and fasted LW at the beginning and end of the experiment. Blood, urine and faeces were sampled after 6 weeks and at the end of the experiment. Jugular blood samples were obtained using vacutainers containing lithium heparin as an anticoagulant. These samples were immediately placed in iced water, the plasma was separated by centrifugation (3000g for 10 min) and stored frozen. Urine samples (~50 mL) were obtained following manual stimulation of the vulva to initiate urination; samples were obtained from 97% of the heifers. The pH of the urine was measured, the urine was acidified to pH < 4 by addition of 10 M HCl, and the samples

Table 2. Composition and the calculated contents of supplements and measured voluntary intakes of supplement components (Experiment 1)

Phosphoric acid was included in all the supplements. Supplement intake is the mean of weeks 2–8 of supplementation. Non-protein nitrogen (NPN) was derived from both the urea and the ammonium sulfate. The NPN in excess of that required to ferment the molasses (g N/day) was calculated assuming that 14 g of rumen-degradable N was required to ferment each kg molasses, and ingested urea N was used with an efficiency of 0.8. LW, liveweight. Values within a row followed by the same letter are not significantly different. *, $P < 0.05$; ***, $P < 0.001$; n.s., not significant

Measurement	Supplement				s.e.m.	Significance
	M80U	M140U	M200U	M140U+AS		
	<i>Ingredients as-fed (g/kg)</i>					
Molasses	896	838	781	616	–	–
Urea	78	137	195	100	–	–
Ammonium sulfate	0	0	0	120	–	–
Phosphoric acid	26	25	24	19	–	–
Water	0	0	0	145	–	–
	<i>Calculated contents</i>					
DM content (g/kg)	725	742	760	666	–	–
NPN (g N/kg as-fed molasses)	41	76	117	117	–	–
NPN (g N/kg DM)	50	86	120	109	–	–
Phosphorus (g P/kg DM) ^A	10.3	9.4	8.6	7.7	–	–
	<i>Supplement intake</i>					
As-fed (g/day)	1105c	476b	256ab	169a	72	***
DM (g/day)	801c	353b	194ab	112a	52	***
DM (g/kg LW.day)	3.61c	1.62b	0.87ab	0.51a	0.224	***
NPN (g N/day)	40.1c	30.3b	23.8ab	12.2a	4.02	*
NPN (g/kg LW.day)	181c	139b	105ab	55a	18.1	*
P (g P/day)	7.19c	2.85b	1.43ab	0.74a	0.38	***
P (g/kg LW.day)	32.4c	13.1b	6.4ab	3.4a	1.71	***
NPN in excess of that required to ferment the molasses (g N/day)	22.8	23.3	19.8	10.4	2.8	n.s.
Fasted LW gain (kg/day)	0.33	0.29	0.23	0.26	0.025	n.s.

^AIncludes P in both the phosphoric acid and the molasses.

Table 3. Composition and the calculated contents of supplements, measured voluntary intakes of supplement components, urea and inorganic phosphorus in plasma and urine and liveweight (LW) change (Experiment 2)

Microbial nitrogen (N) outflow from the rumen was calculated from the excretion of purine derivatives and by using creatinine as a marker of urine output. Urea N in excess of that required to ferment the molasses (g N/day) was calculated assuming that 14 g of rumen-degradable N was required to ferment each kg of molasses, and ingested urea N was used with an efficiency of 0.8. n.s., not significant; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$

Measurement	Supplement				s.e.m.	Significance
	Nil	M200U	M190U+50PA	M180U+100PA		
	<i>Ingredients (g/kg as-fed)</i>					
Molasses	–	800	760	721	–	–
Urea	–	200	191	182	–	–
Phosphoric acid	–	0	49	97	–	–
	<i>Calculated contents</i>					
DM content (g/kg)	–	768	770	772	–	–
Urea N (g N/kg DM)	–	121	116	110	–	–
Phosphorus (g P/kg DM) ^A	–	0.7	16.1	31.5	–	–
Supplement pH	–	5.8	3.4	2.5	–	–
	<i>Supplement intake</i>					
As-fed (g/day)	–	404b	274a	229a	22	*
DM (g/day)	–	310b	211a	177a	17	*
DM (g/kg LW.day)	–	1.53b	1.03a	0.87a	0.090	*
Urea N (g N/day)	–	38b	24a	20a	2.0	**
Urea (mg N/kg LW.day)	–	186	119	96	10	**
P (g/day)	–	0.2a	3.4b	5.3c	0.11	***
P (mg/kg LW.day)	–	1	16	26	0.5	***
Urea N in excess of that required to ferment the molasses (g N/day)	–	33b	22a	17a	1.8	**
	<i>Animal measurements</i>					
Microbial N synthesis (g N/day) – 6 weeks	31.6	34.5	34.6	36.6	2.5	n.s.
Microbial N synthesis (mg N/kg LW.day) – 6 weeks	154	166	162	173	12	n.s.
Microbial N synthesis (g N/day) – 15 weeks	21.2	27.4	27.4	25.9	1.9	n.s.
Microbial N synthesis (mg N/kg LW.day) – 15 weeks	109	129	125	120	10	n.s.
Plasma urea (mg N/L) – 6 weeks	42a	110b	66a	55a	11.1	*
Plasma urea (mg N/L) – 15 weeks	41a	117b	68a	53a	8.1	**
Urinary urea excretion (g N/day) – 6 weeks	1.3a	11.8b	6.2ab	3.8a	1.6	*
Urinary urea excretion (g N/day) – 15 weeks	0.5a	13.1b	4.0a	3.0a	1.2	**
Plasma inorganic P (mg P/L) – 6 weeks	59b	46a	68b	67b	3.6	*
Plasma inorganic P (mg P/L) – 15 wk	54b	37a	64c	70c	1.9	***
Urinary inorganic P excretion (mg P/day) – 6 weeks	56	63	81	134	20	n.s.
Urinary inorganic P excretion (mg P/day) – 15 weeks	47	54	113	112	31	n.s.
Urinary volume (L/day) – 6 weeks	3.8	3.8	3.6	3.2	0.61	n.s.
Urinary volume (L/day) – 15 weeks	2.3ab	3.2b	1.5a	2.2ab	0.28	*
Urinary pH – 6 weeks	7.9	7.9	7.9	7.6	0.10	n.s.
Urinary pH – 15 weeks	7.1	7.3	6.4	6.4	0.29	n.s.
Fasted LW gain (kg/day)	0.07a	0.18b	0.28c	0.22bc	0.023	**

^AMolasses contained 1 g P/kg DM.

were placed on ice. A pooled urine sample for each paddock group was prepared on an equal-volume basis from the individual animal samples and stored frozen. Faecal samples were obtained from the rectum; samples were obtained from 92% of the heifers. These faecal samples were dried (at 70°C) and then ground through a 1-mm screen (Cyclotec 1093 Sample Mill, Tecator, Höganäs, Sweden). A pooled faecal sample for each paddock group was prepared on an equal-weight basis from the individual animal samples.

Urea in plasma and urine was analysed by the method of Tiffany *et al.* (1972), which depends on hydrolysis of the urea and determination of the resultant ammonia; thus, any ammonia present in the urine or plasma would have been measured as urea.

Inorganic P in plasma and urine was analysed by the method of Wang *et al.* (1983). Purine derivatives (allantoin and uric acid) and creatinine in urine were measured by HPLC using a modification of the method of Resines *et al.* (1992). Urinary excretion was calculated from the creatinine concentration, assuming a daily creatinine excretion of 0.558 mmol/kg W^{0.75}; this value had been determined by total collection in similar *Bos indicus* cross cattle fed similar forages, and with analyses in the same laboratory (P. W. Kennedy, unpubl. data). Microbial N outflow from the rumen was calculated from the excretion of purine derivatives as described by Chen and Gomes (1992) and assuming that the endogenous purine-derivative excretion of the *Bos indicus* cross heifers was 0.190 mmol/kg W^{0.75}.day (Bowen

et al. 2006). Total N concentration of faeces was determined by a combustion method (Sweeney 1989), and total P concentration colorimetrically following ashing and acid digestion (AOAC 1980).

For NIRS analysis, faecal samples were redried (at 65°C), cooled in a dessicator and scanned (400–2500-nm range) with a monochromator fitted with a spinning-cup module (Foss 6500, NIRSystems Inc., Silver Spring, MD, USA). Chemometric analysis used ISI software (Infrasoft International, Port Matilda, PA, USA). The Coates and Dixon (2008, 2011) and Dixon and Coates (2009) F.NIRS calibration equations were used to predict the total N and non-grass concentrations and DM digestibility (DMD) of the diet, DM intake and total N concentrations of the faeces. In the pasture system used in the present study, the non-grass was expected to comprise predominantly native forbs, legumes and browses. ME intake was calculated from DM intake and diet DMD (CSIRO 2007).

Results

Experiment 1. Addition of urea and ammonium sulfate to molasses supplements

The molasses contained 698 g DM/kg and, on a DM basis (g/kg), 861 organic matter, 12.6 N, 10.5 calcium and 1.3 P. The concentration of NPN from urea and ammonium sulfate was 117 g N/kg as-fed molasses in both the M200U+PA and M140U+AS+PA treatments, but because of the differing concentrations of N in the urea and ammonium sulfate and the addition of water, the NPN concentration per kg of as-fed supplement and per kg supplement DM was greater in the former supplement (Table 2).

The heifers were in good health throughout the experiment. VI of supplement did not change ($P > 0.05$) through the supplementation interval and the coefficient of variation among weeks ranged from 0.14 to 0.98, averaging 0.40. VI of supplement varied widely among the supplement treatments (Table 2). As the urea concentration in the as-fed supplement was increased from 78 to 195 g/kg, VI of supplement decreased ($P < 0.001$) from 1105 to 256 g as-fed/day, or from 3.61 to 0.87 g DM/kg LW.day. Intake of NPN in the supplement decreased ($P < 0.05$) by 42%, from 40.1 to 23.3 g N/day. VI of M140U+AS+PA supplement containing some ammonium sulfate was lower ($P < 0.05$) than that of M140U+PA, and also tended ($P > 0.05$) to be lower than the intake of M200U+PA. The amount of NPN ingested in excess of that required to ferment to molasses in the supplement ranged from 19.8 to 23.3 g N/day for the three supplements containing molasses, urea and phosphoric acid, but tended ($P > 0.05$) to be lower for the M140U+AS+PA supplement (10.4 g N/day). Fasted LW gain did not differ ($P > 0.05$) among the supplementation treatments and was on average 0.28 kg/day.

Experiment 2. Heifers fed no supplement, or molasses-based supplements containing 200 g urea/kg without or with phosphoric acid

Rainfall during the months October to December preceding the experiment exceeded the long-term average, but the rainfall during the 2 months before and also during the experiment was much lower than the long-term average (Table 1). Thus,

although there was a large amount of pasture DM on offer, due to the early commencement of dry-season conditions, the nutritional quality of pasture during the experiment was lower than generally occurs for these months at the study site. The molasses contained 710 g DM/kg and, on a DM basis (g/kg), 871 organic matter, 11.2 N, 9.6 calcium and 1.0 P. Urea N content of the supplements ranged from 110 to 121 g N/kg DM and the P content from 0.7 to 31.5 g P/kg DM (Table 3). The M200U supplement was pH 5.8, and inclusion of phosphoric acid reduced supplement pH to 3.4 and 2.5.

VI of the supplement did not change ($P > 0.05$) within treatment during the supplementation interval and the coefficient of variation of supplement intake among weeks ranged from 0.15 to 0.33. VI of the M200U supplement was 404 g as-fed/day or 1.53 g DM/kg LW.day, and provided 38 g urea N/day. VI of as-fed supplement, and of supplement DM and NPN, were reduced ($P < 0.05$) by 32–37% by inclusion of the lower concentration of phosphoric acid (M190U+50PA) (Table 3). VI of the supplement containing the higher concentration of phosphoric acid (M180U+100PA; 0.87 g DM/kg LW.day) tended ($P > 0.05$) to be lower than that of the M190+50PA (1.03 g DM/kg LW.day).

Plasma urea concentration was 41–42 mg N/L in the unsupplemented heifers and was increased ($P < 0.05$), or tended to be increased, by the provision of each of the supplements (Table 3). In the unsupplemented heifers, urinary urea excretion was 1.3 and 0.5 g N/day after 6 and 15 weeks, respectively, of supplementation and was increased ($P < 0.05$) by the provision of each of the supplements. The amount of NPN provided as urea, in excess of that required to ferment the molasses in the supplement, ranged from 17 to 33 g N/day for the three supplements. Inorganic P concentration in jugular plasma was 54–59 mg P/L in the unsupplemented heifers and was reduced ($P < 0.05$) to 37–46 mg P/L by provision of the M200U supplement. However, inorganic P concentration in jugular plasma was increased ($P < 0.05$), or tended to be increased, by provision of both supplements containing phosphoric acid. Urinary excretion of inorganic P was 47–63 mg P/day in the unsupplemented and M200U-supplemented heifers, and tended ($P > 0.05$) to be increased by the provision of supplements containing phosphoric acid, to 81–134 mg P/day. Urine pH was not affected ($P > 0.05$) by provision of any of the supplements, and this pH was ≥ 6.4 even when the highest concentration of phosphoric acid was ingested. Microbial N synthesis in the unsupplemented heifers was 154 and 109 mg N/kg LW.day after 6 and 15 weeks, respectively, of the experiment, and tended ($P > 0.05$) to be increased by the supplements.

F.NIRS measurements indicated that the diet selected by the unsupplemented heifers contained 8.9 and 5.7 g total N/kg and had a DM digestibility of 516 and 478 g/kg after 6 and 15 weeks, respectively (Table 4). Non-grass comprised 135 and 91 g/kg after 6 and 15 weeks, respectively. Pasture intakes were estimated to be 18.0 g DM/kg LW.day and 129 kJ ME/kg LW.day after 6 weeks, and 14.2 g DM/kg LW.day and 93 kJ ME/kg LW.day after 15 weeks. Efficiency of microbial N production, calculated as the outflow of microbial N from the rumen measured from excretion of purine derivatives (Table 2) and the ME intake measured by F.NIRS, was 1.2 g microbial N/MJ ME intake

Table 4. The concentration (mean and s.d. on a DM basis) of total nitrogen (N), DM digestibility and non-grass in the diet, concentration of N in faeces and DM intake measured from near-infrared spectroscopy analysis of faeces of heifers in the three replicate paddocks and not fed supplements (Experiment 2)

Metabolisable energy (ME) intake, calculated from DM intake and DM digestibility, and faecal P concentration, are also given. Samples were obtained after 6 and 15 weeks of supplementation and samples from individual heifers were pooled within paddocks. LW, liveweight; P, phosphorus

Measurement	Time of sample			
	6 weeks		15 weeks	
	Mean	s.d.	Mean	s.d.
<i>Diet</i>				
Total N (g/kg)	8.9	1.53	5.7	0.34
DM digestibility (g/kg)	516	11	478	13
Non-grass (g/kg)	135	50	91	33
DM intake (g DM/kg LW.day)	18.0	1.47	14.2	0.73
ME intake (kJ ME/kg LW.day)	129	14	93	6
<i>Faeces</i>				
Faecal N (g/kg)	12	1.0	10	0.9
Faecal P (g/kg)	2.0	0.12	1.4	0.06

after both 6 and 15 weeks. The concentration of P in faeces of unsupplemented heifers was 2.0 and 1.4 g P/kg DM after 6 and 15 weeks respectively. Fasted LW gain was 0.07 kg/day in the unsupplemented heifers, and was increased ($P < 0.05$) to 0.18 kg/day by provision of the M200U supplement (Table 3). Provision of the M190U+50PA supplement caused a further increase ($P < 0.05$) in LW gain to 0.28 kg/day, while the LW gain of the heifers fed the M180U+100PA (0.22 kg/day) did not differ significantly from that in the other two supplement treatments.

Discussion

Effects of urea and phosphoric acid on VI of supplement

In the present studies, both increasing the concentration of urea from ~78 to 195 g urea/kg as-fed, and adding phosphoric acid which reduced the supplement from pH 5.8 to pH 2.5, markedly reduced VI of the molasses-based supplement. In Experiment 2, the VI of the supplement was reduced by 0.20 g DM/kg LW.day for each unit decrease in pH of the supplement. This was a much smaller effect of supplement pH on the VI of molasses-urea supplement than what was observed in a previous comparable experiment (Dixon and Hirst 1999) where VI was reduced by 1.3 g DM/kg LW.day for each unit decrease in pH of the supplement. However, in this latter experiment the concentration of urea in the supplement was much lower (50–57 g urea/kg as-fed supplement) and VI of the supplement was much higher at supplement pH 5 (4.9 g supplement DM/kg LW.day); this may explain the difference between the experiments. In addition, in Experiment 1 of the present study, the VI of the supplement was reduced by 0.02 g DM/kg LW.day for each g/kg increase in urea concentration in the as-fed supplement. This compares with declines in VI of 0.025 and 0.07 g DM/kg LW.day for each g/kg increase in urea concentration in molasses reported for

cattle in pens fed tropical grass forage (Ramirez and Sutherland 1971) or chopped sugarcane (Silvestre *et al.* 1977), respectively. The observation in Experiment 1 that the VI of supplement tended to be reduced by replacement of some of the urea in the supplement with ammonium sulfate is in agreement with reports in cattle that ammonium salts had a greater effect than urea to reduce the VI of cereal grain by cattle (Hough *et al.* 1995), and that sulfate in drinking water reduced VI of forages (Hunter *et al.* 2002). However, a disadvantage of the feed-grade ammonium sulfate used in the present study was that it was difficult to dissolve for incorporation into the supplement.

The reductions in the VI of the supplement due to inclusion of urea or reduction in pH likely involved different physiological mechanisms. Although cattle innately dislike the bitter taste of urea (Goatcher and Church 1970; Grovum and Chapman 1988), it seems more likely that the reduction in the VI of the supplement with increasing urea concentration was associated primarily with development of conditioned flavour aversions to the urea (Chalupa *et al.* 1979; Kyriazakis and Oldham 1993; Villalba and Provenza 1997). In contrast, the reduced VI of the molasses-urea supplement due to the addition of phosphoric acid was likely to be due primarily to an innate dislike by the cattle for the sour flavour of the acids (Goatcher and Church 1970). Experiments where the VI of forages has been reduced by addition of acid to the forage and where animals were sham-fed or intra-ruminally infused with acid have indicated that intake was reduced by taste effects rather than a conditioned flavour aversion (L'Estrange and Murphy 1972; L'Estrange and McNamara 1975; Morgan and L'Estrange 1976; Grovum and Chapman 1988).

The heifers consumed up to 1.2 mmol phosphoric acid/kg LW.day from the supplement in each of the experiments. In previous experiments (L'Estrange and Murphy 1972; L'Estrange and McNamara 1975; Morgan and L'Estrange 1976; Cooper *et al.* 1995), up to ~6 times as much hydrochloric or sulfuric acid per kg LW was fed to or infused intra-ruminally into sheep and cattle, and although the VI was reduced, no adverse effects on the health of the animals were reported. The observation that in Experiment 2 the urinary pH in heifers ingesting the highest amount of phosphoric acid was within the normal range for cattle fed high-concentrate diets (Topps *et al.* 1966) and did not differ among treatments, and that liveweight gain was increased by the supplement providing the highest amount of phosphoric acid, provided evidence that in the present study the heifers were not adversely affected by the phosphoric acid ingested in the supplement. The tendency for urinary P excretion to increase when phosphoric acid was included in the supplement was consistent with increased renal excretion of ammonium and phosphate ions to maintain cation-anion balance during mild acidosis, such as occurs with high-grain diets (Scott 1975), but the amount excreted (<2% of the supplementary P) was not of consequence for the efficiency of utilisation of the supplementary P. There was likely little effect in the rumen of the acid ingested in the supplement, given the high buffering capacity of rumen digesta against acids (Emmanuel *et al.* 1969; L'Estrange and Murphy 1972).

Several studies have shown that the inclusion of phosphoric acid in molasses supplements reduces the risk of urea toxicity (Perez *et al.* 1967; Hemingway *et al.* 1972). Davidovich *et al.* (1977) concluded that inclusion of even 3% phosphoric acid in

a diet allowed urea concentration to be increased substantially without increasing the risk of urea toxicity; this occurred because the acid prevented an increase in rumen pH following ingestion of supplement containing urea and thus prevented rapid absorption of the ammonia derived from the urea, which causes urea toxicity. This is similar to the situation in cattle fed high-grain diets where high concentrations of urea can be fed without risk of urea toxicity. Apart from reducing the VI of the supplement and providing supplementary P, the inclusion of phosphoric acid in molasses–urea supplements substantially reduces the risk of urea toxicity when high concentrations of urea are included.

Responses by the heifers to the supplements

The increase in LW gain from 0.07 to 0.18 kg/day (i.e. 3.5 kg/month) due to provision of the M200U supplement in Experiment 2 was in accord with the increased LW gain, or reduced LW loss, due to provision of molasses–urea supplements in numerous experiments in similar environments (Winks *et al.* 1979; Dixon and Doyle 1996; Dixon 2011). The N and ME concentrations of the diet selected, the intake of ME and microbial N synthesis as measured with F.NIRS and by excretion of purine derivatives, were comparable with previous measurements of cattle grazing speargrass native pasture in the same region (Dixon *et al.* 1998, 2011a, 2011b). The absence of a LW benefit to providing molasses–urea supplement in Experiment 1 was likely associated with the higher rainfall and thus higher pasture quality, and the short supplementation interval.

The concentration of inorganic P in plasma from the jugular vein of M200U-supplemented heifers in Experiment 2 (37 and 46 mg P/L) indicated that a marginal P deficiency was likely (Wadsworth *et al.* 1990). The concentrations of P in faeces of 2.0 and 1.4 g P/kg DM in unsupplemented heifers indicated that the P concentration in the diet was ~0.93 and 0.62 g P/kg DM, respectively, after 6 and 15 weeks (Dixon and Coates 2011); this diet P concentration was lower than that (~1.3 g/kg DM) required for adequacy of these heifers in slow growth (CSIRO 2007). This is consistent with the measured increase in LW gain due to supplementary phosphoric acid, which confirmed that these heifers were deficient in P, at least when N and ME intakes had been increased by the molasses–urea supplement. Previous experiments with young cattle grazing similar pastures at the same experimental site have reported LW-gain responses to P supplements in several experiments where molasses–urea–phosphoric acid supplements were fed using roller drums (Winks 1990). Adverse effects of phosphoric acid have been observed in some experiments where fertiliser grade ‘black phosphoric acid’ was used as the source of P (Winks *et al.* 1976, 1979). However, McMeniman (1973) reported toxic effects of this latter source of P due likely to the presence of sulfuric acid as a contaminant. Sulfuric acid has much greater effects than the equivalent amounts of phosphoric or hydrochloric acids in inducing metabolic acidosis and reducing forage intake in ruminants (L’Estrange *et al.* 1969; L’Estrange and Murphy 1972). Thus, the adverse effects sometimes observed with ‘black phosphoric acid’ supplements were apparently due to problems with this source of P, and are not likely to occur with technical-grade phosphoric acid.

Modifying supplement mixtures to provide targeted amounts of NPN or P

Clearly, minimum-cost systems to deliver NPN and minerals such as P to grazing cattle must include capacity to provide optimal target amounts of supplement, especially of the higher-cost constituents such as the NPN and P. A feature of the ‘M8U’ (molasses containing ~80 g urea/kg) supplement-delivery system which is widely used in northern Australia (Wythes and Ernst 1984), and also of the comparable supplement containing higher concentrations of urea such as the M200U supplement used in the present study, is that the amount of urea NPN ingested is substantially in excess of that required to ferment the molasses in the supplement and to also provide an appropriate amount of NPN for low-protein dry-season pastures. In the present studies, the amount of urea NPN in excess of that needed to ferment the molasses in the supplement was 33 g N when the M200U supplement was fed, and in the range 17–23 g NPN when phosphoric acid was included in the supplement. It was reduced to 10 g NPN when ammonium sulfate was also included in Experiment 1. Thus, the present study indicated that inclusion of phosphoric acid or ammonium sulfate in molasses supplements allows control of the VI of the supplement and avoidance of excessive VI of NPN. Because of the rapid absorption of ammonia from the rumen of cattle consuming forage diets, some NPN in excess of the requirement calculated from feeding standards (CSIRO 2007) is likely to be desirable to attempt to maintain rumen ammonia supply throughout the 24-h cycle (Dixon 1999). The supplementary NPN required has been estimated directly in field experiments to be ~14 g NPN for the class of cattle used in the present study (Winks *et al.* 1972, 1979). Similarly, manipulation of the concentrations of urea and of phosphoric acid (or an alternative P source) in molasses–urea supplements should allow target VI of supplementary P, such as in the range of 1–5 g P/day, to be achieved using molasses-based supplements.

Insufficient information is available from the present studies on the feeding of molasses-based supplements containing high concentrations of urea and strong acids, to recommend their general use in commercial cattle-production systems. Although no symptoms of urea toxicity were observed in any of the present experiments, numerous differences such as those associated with the behaviour of larger groups of cattle under extensive grazing conditions might introduce problems with the use of molasses supplements containing high concentrations of urea. Indeed, molasses–urea supplements containing high concentrations of urea but without acid (e.g. M200U) were found to be not satisfactory for cattle under extensive grazing conditions, with some occurrences of urea toxicity (D. J. Hirst and R. M. Dixon, unpubl. obs.). Further experimentation and experience is required to understand where such molasses–urea–acid-based supplements can be used satisfactorily in large herds and on commercial properties.

Conclusions

The present studies extended knowledge of the manipulation of VI of molasses-based supplements containing a range of concentrations of urea and phosphoric acid. These observations provide the information necessary to manipulate

these concentrations in molasses–urea supplements to achieve target intakes of supplementary NPN and P. However, further information is needed on the likelihood of urea toxicity of these supplements to cattle in extensive grazing situations.

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