GROWTH AND GLUCOSE METABOLISM IN YOUNG CALVES GRAZING TROPICAL PASTURE - THE EFFECTS OF SUPPLEMENTATION WITH MAIZE OR COTTONSEED CAKE

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Dairy calves (10 weeks old) were allowed to graze irrigated kazungulu setaria pasture Five groups of four calves were each given 1 kg/d of a concentrate supplement made up of a maize and cottonseed meal mixture. The ratio of maize to cottonseed meal was 6:0, 5:1, 4:2, 3:3 or 0:6 to give 15, 23, 31, 39 and 63 gN/kg OM in the supplement respectively. The sixth group was an unsupplemented control. Supplementation resulted in a large increase in the rate of live weight gain (243 v 561 g/d) but there was no difference in the mean liveweight gain of any of the supplemented groups. The conversion of supplement to liveweight gain indicated that on this pasture (despite the availability of excess pasture) the calves responded to the extra energy intake from the supplement. Glucose entry rates were increased in all calves by supplementation. This immediate increase in glucose entry rate together with the significant relationship between glucose entry rate and liveweight gain indicates the potential usefulness of this measurement in predicting (over a short time period) the likely responses to supplementation of cattle at pasture.

Key words: Dairy calves, irrigated setaria pastures, liveweight gain, supplementation, maize, cottonseed meal, glucose entry rates

The growth of weaner calves on tropical pastures is often low due apparently to low pasture intakes. However, the growth of calves allowed to suckle their dams for a short period of time after milking can be extremely high even with only small inputs of milk of about 2 1/2 litres/day (Alvarez, 1980). Milk provides nutrients postruminally (ie they bypass the rumen) through activation of the oesophageal groove reflex. Whilst restricted suckling appears to be the most efficient way to supplement such young calves it is not accepted in 'technologically advanced' countries. Dairy calves must be raised to provide herd replacements yet the required level of supplementary feeding has not been determined. In the study presented here the effect of using two supplements or combinations of these, which have a potential for supplying nutrients postruminally, on production of dairy calves on irrigated tropical pasture was studied. There is evidence that gluconeogenesis in ruminants is controlled by quality and quantity of the feed given (Judson and Leng 1968, 1973; Reilly and Ford 1971); and glucose entry rates have been investigated in adult ruminants as a measure of the adequacy of various dietary regimes (Leng 1970; Bergman 1973). Glucose entry rates are also related to growth rate (Smith et al 1979) in cattle suggesting that they

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may be an aid to predicting the nutritional status and potential growth rate of calves and hence indicate optimal supplementation rate. For these reasons glucose entry rates were measured in dairy calves grazing setaria pasture and receiving isocaloric supplements having different protein content.

Materials and Methods

Location: The experiment was conducted between September and December 1977, at the State Department of Primary Industries Dairy Research Station, Ayr, situated 85 km south of Townsville, at about 19°S latitude.

Experimental animals and pasture: Twenty-four Friesian bull calves reared on milk replacement diets to 10 weeks of age were weaned and transferred to the irrigated pastures.

The pasture, kazungulu setaria (*Setaria anceps* var. kazungulu), had been grazed with adult animals to remove excess feed and it had been given 10 days without grazing before the calves were introduced. The pasture was divided into twelve equal sized areas containing a trough for water. The stocking rate was one calf per 0.1 hectare and the calves entered on short, leafy pasture. me pasture was fertilized regularly and received dressings of nitrogen, 3 kg/ha , every 6 weeks. One dressing of superphosphate (43 kg/ha) and potash (62 kg/ha) was applied immediately before the experiment. The pastures were spray irrigated every three weeks. Pasture samples for analysis were obtained by clipping from randomized quadrates at several times during the experimental period.

Experimental design: A randomized block design with two replicates within each treatment and two animals within each replicate was used. The twenty-four calves were allocated to the six treatment groups by stratified randomization on liveweight basis.

Supplementary feeds: The supplements consisted of crushed maize (M) and ground cotton seed meal (CSM), used in different proportions to formulate isocaloric supplements (14 MJ ME/head/day) with different protein contents as shown in Table 1. The calves were fed 1 kg/head/day of supplement.

Treatment	Pr	oportion of:	Nitrogen content
	Maize	Cottonseed meal	(gN/kg OM)
1	0	0	о
2	6	0	15
3	5	1	23
4	4	2	31
5	3	3	39
6 .	0	6	63

Table 1:

The supplements were given once a day at 0600 hours and each animal was fed individually. All animals were allowed free grazing within the paddock and fresh water was available at all times. Liveweights of the calves were recorded weekly.

Measurement of glucose entry rates. Glucose entry rates were measured in all calves two weeks after the start of the trial and again after 12 weeks. A single injection technique was employed using [2-³H] glucose (supplied by the Radiochemical Centre, Amersham, England) (see Judson and Leng 1972). Two measurements were made on each of the two experimental days. The first dose was administered at 0900 hours (3 hours after the supplement was fed), and the second at 1500 hours.

The calves were each dosed with $150 \,\mu$ Ci[2-³H] glucose and 2 mg glucose carrier in 10 ml 0.9% (w/v) NaCl, through a polyethylene catheter (1 mm ID; 1.50 mm OD) which had been placed in the jugular vein the previous afternoon. Blood samples (10 ml) for assay of the specific radioactivity of plasma glucose were taken into heparinized tubes at 30 min intervals for 3 hours after each dosing. Further blood samples were taken at 6 and 18 hours after the second dosing on each day and the blood water was assayed for ³H.

Analytical methods feed analysis: Pasture, maize and cotton seed meal samples were dried at 100°C for 24 hours in a forced draught oven and ground to pass a 1 mm screen in 20 cm laboratory mill. Total N content in these samples was determined by microkjeldahl digestion, steam distillation and titration. Organic matter was determined following ashing at 600°C.

Plasma glucose, urea and plasma concentration and specific radioactivity. Blood samples were centrifuged and the plasma decanted and stored at -15°C until analysed. Plasma urea concentrations were determined by the diacetyl monoxime method (Marsh et al 1965). Plasma was deproteinized with 2.9% perchloric acid and glucose was determined by the glucose oxidase method of Huggett and Nixon (1957). Glucose was isolated as the pentacetate derivitive (Jones, 1965) and assayed for radioactivity in a Packard, Tricarb liquid scintillation spectrometer (Model 3320).

Body water determination: Body water was obtained from plasma by vacuum sublimation and assayed for radioactivity by liquid scintillation counting (Bray, 1960). Total body water was calculated from the exponential loss of radioactivity from plasma water, assuming that all the ³H from [2-³H] glucose was lost to water within the body (Judson and Leng 1972). The time of administration of the second dose each day was taken as zero time. Total weight gain minus water was assumed to be real tissue gain.

Statistical Analyses: The effects of treatments, time, replicates and interactions on glucose entry rates and plasma glucose concentrations were analysed by the split plot analysis of variance, with unequal sample sizes by the least squares method. The effects of treatments and replicates on liveweight gain and tissue gain were determined by nested analysis of variance with unequal sample sizes (Sokal and Rohlf 1968). The plasma urea concentrations data were subjected to analysis of variance for treatment effects. The plasma urea concentrations were regressed on level of protein supplementation for both the periods, for relationship between these two parameters.

To compare the supplemented groups with the controls and all treatments between themselves, all data were analysed using a priori tests of significance (Sokal and Rohlf 1968).

Results

Pasture quality: The pasture dry matter on offer appeared to be adequate at all times. The mean availability was 3657 kg/ha (range 619-11960) containing 14.3 gN/kg OM (range 8.4-29.4).

The crude protein content of pasture declined rapidly during the last six weeks, but was always above 10 g N/kg OM.

Liveweight change and body water. All calves increased in liveweight during the 14 week experimental period; however, those animals receiving supplements gained more weight (P<0.05) than the unsupplemented control group (Table 2). There was no significant difference between the supplemented groups.

Toble 2:

Mean (+ SE) liveweight gain (g/d) and mean (+ SE) tissue gain (g/d) by calves grazing setaria pasture and receiving isocaloric supplements of different nitrogen content (g/kg DM) for 14 weeks. Four animals were used per treatment, the control group was not supplemented

Treatment	¢	Proportion of:		Initial	Final	Liveweight	Tissue	
		Maize	CSM1	(kg)	(kg)	(g/d)	(g/d)	
1		0	0	58	89	243 + 43 ^{a*}	$34 \pm 17^{a^*}$	
2.		6	o	58	128	575 ± 51^{b}	142 ± 29^{b}	
3		5	1	57	113	505 ± 113^{D}	147 ± 62^{D}	
4		4	2	56	127	570 ± 43^{D}	$71 \pm 34^{\circ}$	
5		3	3	57	1 31	628 ± 84 ^b	137 ± 55	
6		0	6	57	121	**527 ± 25 ^D	$130 \pm 34^{\circ}$	

Values within any one column with different superscripts differ significantly (P<0.05)

Mean of three animals

Cottonseed meal .

Values for body water were extremely variable (CV 20%). Means for tissue gain by the animals in each treatment group are shown in Table 2. There was no significant difference between the supplemented groups in tissue gain, however, animals in these groups produced more tissue dry matter during the experimental period than the control calves (P<0.05).

Plasma glucose concentrations and glucose entry rates Table 3 shows plasma glucose concentrations and glucose entry rates of the calves after 2 and 14 weeks on the experimental treatments (12 and 24 weeks of age respectively). Plasma glucose concentrations of the supplemented groups were always higher than the control groups (Table 3), although this difference was not significant for treatment 5 during period I, or treatment 3 during period II. There was no significant difference between the supplemented groups in plasma glucose concentrations.

Table 3:

Mean (+SE) plasma glucose concentration (mg/litre) and mean (+SE) glucose entry rates (mg/min) for groups of young calves grazing setaria pasture after 2 weeks (Period 1) and 14 weeks (Period II) receiving isocaloric supplements of different nitrogen contents (g/kg OM). Four animals were used per treatment; the control group was not supplemented.

Trestment	Proportion of:		Period	<u> </u>	Period II		
Ireatment	haize	CSM 1	*Plasma glucose concentration (mg/l)	<pre>#Glucose entry rate (mg/min)</pre>	*Plasma glucose concentration (mg/1)	<pre>#Glucose entry rate (mg/min)</pre>	
1	0	0	480 + 15 ^{a**}	125 + 7 ^{a**}	549 + 24 ^{a**}	319 + 68 ^{a*}	
2	6	0	731 + 25 ^b	217 + 45 ^b	$17 + 32^{b}$	$472 + 83^{b}$	
3	5	1	627 ± 21^{b}	198 + 29 ^b	668 + 27 ^a	495 + 79 ^a	
4	4	2	678 ± 31^{b}	191 + 37 ^b	716 + 32 ^b	482 + 101 ^b	
5	3	3	582 + 17 ^a	176 + 28 ^b	$695 + 19^{b}$	553 + 115 ^b	
6	0	6	656 ± 24^{b}	$207 + 23^{b}$	$736 + 18^{b}$	507 + 44 ^b	
	-					0 -1 0	

Cottonseed meal

Mean of 13 samples collected over 9 hours

** Value within any one column with different superscripts differ significantly (P<0.05)

Mean of two measurements; one in the morning and one in the afternoon on the same day

Glucose entry rates increased in all animals with age. Measurements of glucose entry rates made during the morning (mean \pm SE of 177 \pm 10 and 447 \pm 16 mg glucose/min for periods I and II respectively), were close to those during the afternoon (mean \pm SE of 193 \pm 11 and 497 \pm 35 mg glucose/min for periods I and II respectively), and were not significantly different. Animals receiving supplements had higher glucose entry rates (P<0.05) than the unsupplemented control calves (Table 3) and the relationship between supplemental protein intake and glucose entry rates is shown in Figure 1.

Relationship between glucose entry rates and liveweight gain Liveweight gain during the 14 week experimental period was positively correlated (r = 0.87) with glucose entry rates measured at the end of the experiment. Figure 2 shows the relationship between liveweight gain and glucose entry rates. The regression equation describing this relationship was:

 $Y = 1.3 (\pm 0.17) X - 136 (\pm 83)$: RSD = 90.....(1)

where Y is liveweight gain (g/head/day), X is glucose entry rate (mg/min) and RSD is residual standard deviation.

Plasma urea: Table 4 shows the plasma urea concentrations of animals in the 6 treatment groups. The relationship between supplemental protein intake and blood urea concentrations in the 12 and 24 week old calves after 2 and 14 weeks supplementation respectively, are shown in Figure 3. Plasma urea concentrations (Yu) were positively correlated with level of protein supplementation (P) during both measurement periods (r = 0.96 and 0.82 for periods I and II respectively), however, the regression coefficients differed significantly (P<0.01) between periods.

$Yu = 2.82 (\pm 0.94) + 0.025 (\pm 0.04)P$	RSD = 1.82	Period 2	 (2)
$Yu = 0.16 (\pm 0.83) + 0.052 (\pm 0.003)P$	RSD = 1.58	Period 1	 (3)

Plasma urea concentrations were not correlated significantly with glucose entry rates; the relationships are shown in Figure 4.

Table 4:

Mean (+SE) plasma urea concentration (mg/100 ml) for groups of young calves grazing setaria pasture after 2 weeks (Period I) and 14 weeks (Period II) and receiving isocaloric supplements of different nitrogen contents (g/kg DM). Four animals were used per treatment; control group was not supplemented

Treatm	ent Proportion of		Nitrogen content		Plasma u	rea
	Maize	CSM ¹	of supplements control		Period I	Period II
1	0	0	Control		13.2 + 3.7 ^{a*}	$6.8 + 0.4^{a^*}$
2	6	0	15.3		$3.8 + 0.5^{b}$	4.8 ± 0.5^{a}
3	5	1	23.3	¥2	6.5 + 0.6 ^{bc}	$5.0 + 0.4^{8}$
4	4	2	31.2		$9.6 + 0.4^{\circ}$	9.4 ± 1.4^{b}
5	3	3	39.2		13.0 ± 0.6^{a}	8.6 ± 0.7^{b}
6	0	6	63.1		$18.0 + 1.2^{d}$	$11.7 + 0.4^{\circ}$

Cottonseed meal

Values within any one column with different superscripts differ significantly (P<0.05)

Discussion

The results of the feeding trial clearly indicate the low nutritional value of this tropical pasture for early weaned calves. This is apparent even though the availability of pasture was always in excess of the animals intake and it was always in a young vegetative state due to the management practices. Although tropical grasses are generally lower in digestibility than temperate pastures it is difficult to accept that it was the digestibility of the pasture that was the major limitation and which set the upper limit to production in the absence of supplementation. The pasture was however always relatively low in protein (8.4 - 29,4; mean 14.3 g N/kg OM on offer) which is possibly marginal for efficient rumen fermentation and if all the protein was fermented in the rumen it is unlikely that this pasture could support high growth rates (0rskov 1970).

The two supplements were selected because they both have properties which tend to allow them to bypass the rumen and supply bypass starch (maize) (Armstrong and Smithard 1979) ; and bypass protein (cottonseed meal) to ruminants (Kempton et al 1977). The response of ruminants to bypass protein on a diet low in dietary bypass protein is to increase feed intake (Kempton et al 1977) ; the response to bypass starch appears to increase the efficiency of feed utilisation without effecting feed intake (Preston and Leng 1980); the effect of supplementation with material which is rumen fermentable is usually to reduce the ad libitum intake of the basal diet. In these studies

Figure 1:

Effect of varying concentrations of protein in isocaloric supplement (1 kg/head/d) on the glucose entry rates in calves (°), 12 weeks old; (•), 24 weeks old), grazing setaria pasture. Four animals were used per treatment. Control groups (period I), -#-#-; period II, ----) received no supplement



Figure 2:

Relation between glucose entry rates and liveweight change of calves grazing setaria pasture and receiving isocaloric supplements of different protein contents



Figure 3:

Relation between crude protein intake and plasma urea concentration of calves (°), 12 weeks old; (•), 24 weeks old), grazing setaria pasture and receiving isocaloric supplements. Four animals were used per treatment. Values for plasma urea concentration for unsupplement calves were 13 and 17 mg/100 ml for periods I and II respectively



Figure 4:

Relation between plasma urea concentrations and glucose entry rates in calves (°), 12 weeks old; (•), 24 weeks old), grazing setaria pasture and receiving isocaloric supplements of different protein contents. Four animals were used per treatment



supplementation gave liveweight increase of about 250 - 275 g/head per day which is a conversion rate of supplement to bodyweight gain of about 4:1 indicating that the supplement probably partially replaced the basal feed (which was pasture) and the major response therefore was to increase the overall digestible energy content of the diet. The extent of fermentation in the rumen of the supplements was! not measured, however the use of cottonseed meal on low quality roughage diets (adequate in fermentable N through urea supplementation) has been to increase feed intake substantially indicating its bypass nature (Hennessey 1980). However the growth rates of calves obtained in these studies with supplementation are close to maximum expected for the age and the size of the calves and similar to those obtained with calves that are restricted suckled (Alvarez 1980).

The results of this experiment are in agreement with previous observations, which showed that gluconeogenesis was dependent on digestible organic matter intake (Judson and Leng 1968; Steel and Leng 1973 b; Smith et al 1979) as no difference was noted in the calves receiving the isocaloric supplements despite their different content of crude protein. Glucose entry rates increased with growth, which could be attributed to higher feed intake. The linear relationship between glucose entry rates and liveweight change was also consistent with previous findings in lambs and cattle (Kempton and Leng 1980; Smith et al 1979). Plasma glucose concentrations of the supplemented groups were always higher than the control group, being similar to those reported for sheep by Steel and Leng (1973a).

It has been well established that the availability of propionate is a major determinant of rate of synthesis of glucose in fed ruminants (Steel and Leng 1973b). While glycerol may also be an important glucose precursor in the underfed animal (Bergman et al 1968), protein is regarded as being the most likely alternative source of significant quantities of glucose in well fed ruminants (see Leng 1970; Bergman 1973). Estimates of glucose carbon derived from plasma amino acids range from 11 to 30% in fed ruminants (Black et al 1968; Ford and Reilly 1969, 1970; Reilly and Ford 1971; Wolff and Bergman 1972; Bergman et al 1974). However, in view of the present results and suggestions of Wolff et al (1972) it appears that protein carbon can only be utilized to a minor extent. This agrees with the suggestion that gluconeogenesis in cattle on molasses based diets is entirely via an initial reaction involving carbon dioxide fixation (Nagy and Leng 1980). The linear relationship between protein intake and glucose entry rates in sheep pregnant with twins, noted by Reilly and Ford (1971) would indirect! indicate the level of energy intake, as the rations they used increased in energy content with each increase in protein level. Wastage of protein in ruminants on high protein diets has been demonstrated by Wolff et al (1972), who found that as much as 48% of the total dietary nitrogen would be converted to ammonia in the portal drained viscera.

Plasma urea may arise as a metabolic end-product of protein catabolism, or as the result of fermentation of soluble protein in the rumen resulting in increased ruminal fluid ammonia and plasma urea concentrations (Nolan and Leng 1972; Wolff et al 1972). No significant relationship between plasma urea concentrations and glucose synthesis rate was noted in the present study, being consistent with the findings of Leonard et al (1977).

The plasma urea concentrations in high protein supplemented groups and the control group during period II were significantly lower than period I; this could be attributed to either better nitrogen utilisation by ruminal microbes or less availability of protein due to declining pasture quality. Stobo and Roy (1973) emphasized the correct balancing of energy and protein contents of diets for growing calves and demonstrated that crude protein content could be reduced to 153 g/kg DM, for calves of 122 kg liveweight to achieve weight gains of 1 kg/d. Provided the pastures were of sufficient quality (crude protein about 5.2%). No advantage was obtained in the present study from using an isocaloric supplement containing more than 89 g of crude protein per kg DM (in which there is considerable bypass protein) for calves weighing up to 103 kg and gaining weight of 0.58 kg/d.

Egan and Black (1968) and Black et al (1968) suggested that amino acids released from protein are utilized so that there is a prolonged availability of glucose precursors, and the animal can form glucose over longer intervals. This in turn ensures more efficient protein utilization for glucogenic precursors. According to the observations of Sutton (1971) there should be more flux of glucogenic precursors 3 to 4 hours after meals, which was not observed in this experiment in which higher glucose entry rates were noted in the afternoon than the morning.

The linear relationship (r = 0.87) recorded between glucose entry rates and liveweight gains in the present study indicates that measurement of glucose entry rates could be used for prediction of nutritional status and performance, hence the optimal rate of protein and energy supplementation. The present experiment clearly demonstrates that the major requirement of growing calves on tropical pastures is extra energy and glucose entry rate may be an immediate indication of the benefit of a supplement to calves at pasture. Previous recommendations of protein requirement of calves require reconsideration, as from this study protein requirements of calves at pasture may be much less than present recommendations, provided supplements are used which have potential for bypassing rumen fermentation.

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