Glucose Kinetics for Milk Synthesis in Etawah Crossbred Goats Fed King Grass Silage Prepared with Manure**

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ABSTRACT : A study was carried out to determine glucose kinetics, nutrient balance and milk production of lactating Etawah crossbred goats. The animals (27.2 to 29.1 kg BW) were randomly divided into four levels of dietary treatment groups: the first group R1 received 100% (3 kg) fresh king grass (*Penisetum purpuroides*), the second group R2 received 75% king grass and 25% king grass silage prepared with chicken manure, the third group R3 received 50% king grass and 50% silage, and the fourth group R4 received 100% silage. In addition to the roughage, each group received 800 g of concentrate (CP 14.77% of DM; 17.26 MJ/kg). Animals fed king grass silage made with chicken manure were found to be superior to the group fed king grass alone. Glucose kinetics and retained energy were significantly affected. Calculations showed that glucose requirements for maintenance and milk production can be met for the groups with high levels of silage (R3 and R4). The values of glucose flux were in the range of 2.52 to 4.50 mg/min.kg $BW^{0.807}$ which are lower, but close to, the values for the temperate lactating dairy cow. The present glucose flux value for the lactating Etawah crossbred goat is higher than the previous value published from this laboratory. ***(Asian-Aust. J. Anim. Sci. 2002. Vol 15, No. 7 : 982-985)***

Key Words : Etawah Crossbred Goats, Energy and Protein Requirements, Glucose Kinetics, Lactation, Silage

INTRODUCTION

Milk of tropical ruminants such as the Etawah crossbreed goat (Astuti et al., 2000) and the Bali cow (Sukarini et al., 2001) have a striking high content of fat, 6 to 8% as compared to 3.5 to 4% for many temperate dairy breeds. The high fat content is accompanied by low volume of milk. However, high fat content of milk may provide adequate energy for offspring to survive. This could be of advantage to the offspring born in the dry season where adequate forage is not available for the young animal. Inadequate nutrient intake results in retarded growth of the young leading to high mortality during the pre-weaning period. Furthermore there is also the situation in tropical countries that feeding grass alone results in a low level of animal productivity. To increase the lactating performance of tropical ruminants, the degradability of the roughage must be improved.

Basic research on the role of glucose in lactating ruminants is very important because glucose plays a crucial role in their intermediary metabolism, e.g. in mammary metabolism (Chaiyabutr et al., 2000) for the biosynthesis of lactose, fat, regulation of milk secretion (Mepham, 1993) and for many other functions. In the publication on goats from this Institute cited above (Astuti et al., 2000), it was reported that glucose flux in Etawah crossbred lactating goats is low compared to that of temperate ruminant breeds which may implicate the role of glucose to support production, e.g. for NADPH formation required for milk fat fatty acid synthesis. Cross breeding between the Etawah goat which is regarded as a dairy goat, and the indigenous Kacang goat reared for meat, both breeds being regarded as indigenous to Southeast Asia (Devendra, 1993), is common and a variety of genetic combinations exists. One can relate level of milk production of crossbred Etawah phenotypically to the length of the draped ear, this being longer if the animal is genetically closer to the Etawah breed. The opposite, short upright ears, indicate closeness to the Kacang. The low glucose flux values reported by Astuti et al. (2000) may have been because the goats used in their experiments were genetically closer to the Kacang. It is thus of interest to obtain glucose flux values of crossbred goats closer to the Etawah and investigate if the values resemble more closely to those of other dairy ruminant breeds.

The present research on lactating crossbred goats, anticipated to have more Etawah carrying traits, gives special attention to glucose kinetics in general by treating the animals with several different levels of feeds rich in glucogenic precursors. Abundant level of precursor for the

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synthesis of glucose, namely propionic acid, will promote a higher level of lactose synthesis which would augment milk yield compared with a low level of precursor. Another aspect of importance of glucose metabolism is its supporting role in fat formation by supplying NADPH for long chain fatty acid synthesis in the lactating mammary gland. Ensiling grass with chicken manure is expected to preserve as well as improve the quality of animal feed, especially in the dry season. Data on energy and nitrogen balances have been presented previously (Kiranadi et al., 1994).

MATERIALS AND METHODS

Animals and feeding

Twenty Etawah crossbred lactating goats having draped ears of at least 20 cm long, presumed to indicate closeness to the purebred dairy Etawah, and weighing around 27 to 29 kg were randomly divided into four dietary treatment groups with five replications. The animals were previously synchronized for breeding thereby resulting in uniformity of their lactational condition. The animals were in the seventh week of their lactation. The dietary treatments were 4 levels of silage in addition to 800 g of concentrate per day given to each animal. The first group (Rl: BW±SD=27.2±0.5 kg) received 3 kg king grass (100%), the second (R2: 27.8±0.3 kg) 75% king grass and 25% silage, the third (R3: 28.3± 0.5 kg) 50% king grass and 50% silage and the fourth group (R4: 29.1±0.6 kg) received 100% silage. The proximate nutrient contents of the feeding stuffs are shown in table 1. Balance trials were conducted for 14 days adaptation with one week collection. The goats were milked twice daily and the milk of each goat during the collection was pooled to record its production and composition. Milk samples were analyzed for fat and protein by standard chemical milk analysis, while energy content of fat and protein were calculated using values of 38.50 kJ/g for fat and 23.85 kJ/g for protein. Lactose was assumed to be 4.6% of milk and its energy content was estimated as 16.74 kJ/g. Balance of nutrients was calculated from the digestibility and metabolic data. Digestible energy (DE), metabolizable energy (ME), retained energy and retained protein were calculated from the balance. ME values were corrected for rumen CH_4, assumed to be 10% of feed gross energy. Energy in urine was calculated from urinary-N (g/day) times 6.25 times 5.02 kJ. All data were evaluated using analysis of variance and the difference between means was subjected to Duncan Multiple Range Test (Steel and Torrie, 1986).

Table 1. Nutrient contents of feeding stuffs

	King grass	Silage	Concentrate
Dry matter (DM, % of fresh)	35.88	22.52	90.55
		% of DM	
Crude protein	9.37	5.11	14.77
Crude fibre (CF)	38.99	40.99	10.61
Fat	2.20	2.31	8.55
N-free extract	39.02	38.05	50.25
Ash	10.42	13.54	6.37
Calcium (Ca)	0.28	0.44	0.86
Phosphorus (P)	0.22	0.27	0.71
Energy, (MJ/kg)	18.20	16.80	17.26

Glucose kinetics

Glucose kinetics studies were carried out using the pulse labeling technique of glucose-2-^{3}H as has been described previously (Sastradipradja et al., 1976, 1992). Each animal received intravenously 100 μCi labeled glucose and serial blood samples were withdrawn from the jugular vein via implanted catheters at 0 min. and at every 20 mins post injection up to 120 mins. Blood samples (10 ml) were immediately transferred to a centrifuge tube containing 5 mg sodium fluoride and 2 drops of heparin (10 IU/ml) and placed on ice to chill. The blood samples were then centrifuged to separate the plasma from the blood corpuscular elements. The plasma was subsequently transferred into capped plastic tubes and stored frozen at -20°C until assay for glucose concentration and radioactivity. The glucose penta-acetate derivative method (Jones, 1965) was used for measuring plasma tritiated glucose specific activity (SA) for calculation of glucose pool, flux and space of distribution. The glucose flux was calculated by multiplying the glucose pool by the disappearance coefficient found from a straight line fitted to the logarithm transformed glucose SA data collected between 20 and 120 mins. Flux=Pool×k; k=disappearance coefficient of specific activity with time derived from the equation $SA_t=SA_0e^{-kt}$; Pool=(single injected dose of label compound)/(extrapolated SA of label at zero time).

The transfer quotient which is the ratio between the SA of glucose-^{14}C and that of bicarbonate-^{14}C, as an index of gluconeogenesis involving fixation of CO_2 into glucose, and the daily heat production estimated by the carbon dioxide entry rate technique, used primed continuous infusion of approximately 150 μCi of $NaH^{l4}CO_3$ total activity (Manik and Sastradipradja, 1989; Sastradipradja et al., 1991). Because the units of glucose-^{14}C and bicarbonate-^{14}C specific activities were in μCi/at C, gluconeogenesis involving CO_2 fixation was found by multiplying transfer quotient by 6 times the glucose flux.

RESULTS AND DISCUSSION

The results are summarized in table 2. The energy

Table 2. Intakes, digestibilities and metabolism of nutrients, glucose kinetics and milk production of lactating goats

Parameter	R1	R2	R3	R4	SEM	P
Intake						
DM (g/d)	754.28	877.02	999.78	1,081.18		
Energy (MJ/day)	13.55^{b}	15.54^{b}	17.46^{ab}	18.30^{a}	0.29	<0.01
Protein (g/day)	73.12	106.12	127.78	129.98		
Digestibility						
DM (%)	63.05^{b}	64.79^{ab}	67.98^{ab}	75.40^{a}		
Protein (%)	55.88^{b}	66.96^{b}	70.74^{b}	77.43^{a}		<0.05
DE (MJ/day)	8.63^{c}	10.72^{b}	12.6^{ab}	13.92^{a}	0.38	<0.01
Metabolism						
ME (MJ/day)	7.00^{c}	9.47^{b}	10.46^{b}	11.85^{a}	0.209	<0.01
Urinary-N (g/day)	17.16	13.50	22.99	24.10		
Urinary E (MJ/day)	0.536^{b}	0.424^{b}	0.60^{ab}	0.77^{a}	0.063	<0.01
HP (MJ/day)	4.76	4.11	4.11	4.14		
Retained E (MJ/day)	1.94	5.02	6.05	7.01		
Rprot (g/day)	23.08^{d}	56.95^{c}	67.42^{b}	76.13^{a}	14.02	<0.05
(equivalent to energy MJ/day)	0.55^{d}	1.358^{c}	1.608^{b}	1.816^{a}	0.334	<0.05
Glucose kinetics						
Blood glucose (mg%)	51.52	55.70	53.20	59.34		NS
Pool size (mg/kgBW)	135.95^{c}	135.95^{c}	135.95^{c}	135.95^{c}		NS
Flux (mg/kgBW$^{0.807}$)	2.52^{b}	3.12^{b}	4.10^{a}	4.50^{a}	0.038	<0.05
Transfer quotient= TQ $CO_2 \rightarrow$glu (%)	6.12^{c}	8.40^{b}	10.50^{a}	10.90^{a}		<0.05
Milk production yield (ml/day)	98.4^{b}	101^{b}	85^{b}	154^{a}		<0.05
Fat (%)	3.96^{b}	4.14^{b}	3.74^{b}	6.51^{a}		<0.05
Protein (%)	3.32^{c}	4.25^{d}	4.97^{a}	5.03^{a}		<0.05
Energy (MJ/day)	0.303^{b}	0.34^{b}	0.301^{b}	0.70^{b}	0.005	<0.05

Different letters following values in a horizontal row show significant differences at P value indicated. DM=Dry matter: DE=Digestible energy; ME=Metabolizable energy; HP=Body heat production; Retained energy includes energy in milk; Rprot.=Retained protein which includes protein of milk.

balance of the experiments indicated that responses to R3 and R4 were superior to those of R1 and R2. The DM, GE and protein intakes were found to be better on R3 and R4 indicating that increasing silage in these rations improved feed palatability. Ensiling breaks the ligno-cellulose bonds of king grass which increases digestibility and nutrient utilization, and their metabolism. This conclusion was supported by the data on ME, milk production, retained energy, glucose kinetics and gluconeogenesis involving CO_2 fixation. ME:DE ratio, retained energy and retained protein were also better for R3 and R4. Performance on diet R4 was the best with respect to intake and digestibilities and glucose production rate. From the calculation of milk synthesis based on glucose metabolism, assuming that the physiology of milk production for the goat is similar to that of the dairy cow, the glucose required for milk production can be calculated. Milk production requires glucose for the synthesis of lactose, triglyceride and fat. For the synthesis of lactose where lactose is assumed to be 4.6% of milk, the 98ml/day of milk on R1 produced 4.51 g lactose which required 4.75 g glucose/day.head. Similarly for R2, R3 and R4, glucose requirements were 4.89, 4.12 and 7.46 g/day·head, respectively. For the formation of triglyceride-glycerol, 12% of the mols of fatty acids being synthesized is needed (Boekholdt, 1976). Thus for the synthesis of 3.96% fat of milk on R1, the maximum value of glucose for fat would be 0.47 g/day and for R2, R3 and R4 would be 0.50, 0.38 and 1.19 g/day respectively. Synthesis of milk also utilizes NADPH; 1 kg milk fat needs 49 mols NADPH derived from 730 gram glucose, and maximum glucose for fatty acid synthesis is 73% of the glucose supply (Bockholdt, 1976). Assuming that 50% of the NADPH requirement will be supplied by glucose required for fat synthesis, the amount for R1 would be 2.83 g/day. For R2, R3 and R4 it would be 3.03, 3.15 and 7.26 g/day, respectively. Thus the total amount of glucose needed for milk synthesis is the sum of glucose for lactose, fat and NADPH formation. Calculations for R1, R2, R3 and R4 would be 8.06, 8.30, 7.32 and 15.91 g/day, respectively. Assuming that the non-lactating goat required a flux of 2.86 mg/min/kg$^{0.807}$ (Ballard et al., 1969; Astuti et al., 2000 and Sukarini et al., 2001), glucose requirement for the non lactating goats would be 46.20 g/day.

Summing up, the glucose requirement for milk production of lactating goats for R1, R2, R3 and R4 would be 54.26, 54.5, 53.52 and 62.11 g/day, respectively. The value of glucose flux for R1 was 27×60×24 g/day which was only 38.88 g/day, indicating that feeding with king

grass alone was not sufficient for maintaining the lactating animal. For R2 the glucose produced from the flux was 47.52 g/day, indicating inadequate glucose supply for the animal. For R3 the production rate was 61.34 g/day, and the glucose was adequate for milk production. The R4 produced 66.82 g/day glucose, which was a very satisfactory amount for meeting the glucose requirement for milk production. The result of the glucose flux in this experiment, ranging from 2.52 to 4.50 mg/min. $kg^{0.807}$, is relatively higher than the result reported earlier (Astuti et al., 2000). From the calculations above, glucose is confirmed to be an essential nutrient for the goat, especially during lactation, and also pregnancy. Despite a low concentration of glucose and practically an absence of dietary glucose readily absorbed by the gut, ruminants rely on gluconeogenesis to meet its glucose requirement. The transfer quotient measured in this study served as an index of gluconeogenesis which involves fixation of CO_2-carbon to produce glucose, from precursors such as alanine, lactate, pyruvate and propionate. Among these precursors, propionate has preference in endogenous glucose production by the liver over the other compounds (Brockman, 1993). The results demonstrated that feeding goats with king grass silage supplies favorable amounts of glucose to support milk production. In this experiment the levels of nutrient supplied to all animals were adequate for positive energy and protein balance. It can also be shown that feeding the animals with 100% silage produced retained energy of (7.37-2.66)/39.75, or 118 g/day, in contrast to the group fed 100% king grass which showed fat mobilisation. Considering milk fat values of 3.96 to 6.46%, the demand of NADPH through glucose pentose pathway for fat synthesis was not high. The milk protein concentration of 3.32% to 5.03% indicated also that the animal were not demanding high levels of ATP to provide energy for peptide binding of milk proteins.

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