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Maintenance of plasma branched-chain amino acid concentrations during glucose infusion directs essential amino acids to extra-mammary tissues in lactating dairy cows

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ABSTRACT

The objectives of this study were to investigate the effects of branched-chain AA (BCAA) supplementation when glucose is infused postruminally into lactating dairy cows consuming a diet low in crude protein (CP) and to test the hypothesis that low BCAA concentrations are responsible for the poor stimulation of milk protein yield by glucose. Twelve early-lactation Holstein cows were randomly assigned to 15% and 12% CP diets in a switchback design of 6-wk periods. Cows consuming the 12% CP diet received 96-h continuous jugular infusions of saline and 1 kg/d of glucose with 0, 75, or 150 g/d of BCAA in a Latin square sequence of treatments. Compared with saline, glucose infusion did not affect dry matter intake but increased milk yield by 2.2 kg/d and milk protein and lactose yields by 63 and 151 g/d, respectively. Mammary plasma flow increased 36% during glucose infusion compared with saline infusion, possibly because of a 31% decrease in total acetate plus β-hydroxybutyrate concentrations. Circulating concentrations of total essential AA and BCAA decreased 19 and 31%, respectively, during infusion of glucose, yet net mammary uptakes of AA remained unchanged compared with saline infusion. The addition of 75 and 150 g/d of BCAA to glucose infusions increased arterial concentrations of BCAA to 106 and 149%, respectively, of the concentrations in saline-infused cows, but caused a decrease in concentrations of non-branched-chain essential AA in plasma, as well as their mammary uptakes and milk protein yields. Plasma urea concentration was not affected by BCAA infusion, indicating no change in catabolism of AA. The lack of mammary and catabolic effects leads us to suggest that BCAA exerted their effects on plasma concentrations of the other essential AA by stimulating utilization in skeletal muscle for protein accretion. Results indicate that the glucose effect on milk protein yield was not limited by low BCAA concentrations, and that a stimulation of extra-mammary use of non-branched-chain essential amino acids by BCAA led to a decrease in milk protein yield.
Key words: mammary uptake, branched-chain amino acid, glucose, milk synthesis

INTRODUCTION

Of the total digestible EAA that make it into portal blood from the gastrointestinal tract of the lactating dairy cow, approximately 50% is taken up by the mammary glands and used for milk protein production (Lapierre et al., 2012). The remaining half enters into body protein or is catabolized in the liver to produce glucose and ATP. Provision to the cow of additional glucose, precursors of glucose such as propionate, or euglycemic insulin spares NEAA from entering into gluconeogenesis (Lemosquet et al., 2004; Curtis et al., 2014) and directs EAA into protein of both the muscle and the mammary glands (Clark et al., 1977; Bequette et al., 2002; Rulquin et al., 2004). However, evidence of glucose or glucose precursors effectively stimulating milk protein yield has been inconsistent, as some studies show a positive effect (Rulquin et al., 2004; Raggio et al., 2006a) and other studies do not (Hurtaud et al., 1998; Curtis et al., 2014; Nichols et al., 2016).

Concentrations in plasma of the branched-chain amino acids (**BCAA**) Ile, Leu, and Val decline when glucose or propionate is supplied (Raggio et al., 2006a; Curtis et al., 2014; Nichols et al., 2016), which may contribute to small or absent milk protein yield responses to glucose. Nichols et al. (2016) estimated that approximately 136 g/d of BCAA were diverted into non-mammary tissues as a result of infusing 1 kg/d glucose. When 215 g/d of BCAA was subtracted from 1,475 g/d of MP, milk protein yield decreased 123 g/d (Doelman et al., 2015b), suggesting that a loss of 136 g/d of BCAA is sufficiently large to interfere with the ability of glucose to stimulate milk protein production.

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The present experiment was designed to investigate the effects of BCAA replacement when glucose is infused postruminally into lactating dairy cows consuming a low-CP diet and to test the hypothesis that low BCAA concentrations are responsible for the poor stimulation of milk protein yield by glucose. Circulating concentrations and net mammary uptakes of energy metabolites and AA were measured during infusions of 75 and 150 g/d of BCAA to quantify the supply of postruminal BCAA needed to counteract the decrease in plasma BCAA concentrations elicited by 1 kg/d glucose, and to gather evidence as to which pathways of BCAA utilization are responsible for the loss.

MATERIALS AND METHODS

Animals, Diets, and Feeding Management

All experimental procedures were approved by the Animal Care Committee at the University of Guelph (Guelph, ON, Canada). Twelve Holstein cows in their second lactation began the experiment at 80 ± 22 d of lactation and weighed 700 ± 44.3 kg. Cows were housed in tie stalls, given individual free access to water and feed throughout the study, and milked twice daily at 0500 and 1700 h. Two TMR were formulated at 15% (normal) and 12% (low) CP to meet 105 and 87% of MP requirements and 105 and 101% of ME requirements, respectively, for 36 kg/d of milk (NRC, 2001; Table 1). The mean balances for MP, RDP, and RUP for the normal- and low-CP diets, respectively, were formulated to be 127.5 and −339 g/d, −32.3 and 265.2 g/d, and 143 and −309.7 g/d. Additionally, mean balances for EAA, Met, and Lys for the normal- and low-CP diets, respectively, were formulated at 76 and −143.6 g/d, 0.75 and −10.3 g/d, and 1.2 and 29.5 g/d. The 15% CP (normal) ration represented a characteristic corn alfalfa silage-based diet fed to Ontario dairy cattle for the purpose of establishing normal milk production and protein efficiency values with which to compare responses to glucose and BCAA infusions on the 12% CP (low) TMR. Cows were fed at 1200 h daily, and feed offered and refused was weighed to determine daily ad libitum feed intakes of individual cows. Samples of each ration were collected daily, stored at −20°C, and pooled every other week over the 16-wk study for nutrient composition, which was analyzed by wet chemistry at a local commercial laboratory (Agri-Food Labs, Guelph, ON, Canada). Feed refusal samples were collected daily from individual cows, stored at −20°C, and pooled weekly. Dry matter contents of feed and refusal samples were determined using a forced-air oven at 55°C for 48 h.

Experimental Design, Catheterizations, and Jugular Infusions

Cows were assigned the normal- and low-CP diets in a switchback design of two 6-wk periods. The first 2 wk were allocated for diet adaptation, and cows assigned to the low-CP diet were subjected to a series of infusions for the remaining 4 wk. Each of these 4 wk consisted of 96 h of continuous infusion, followed by 72 h of washout with no infusion, in a randomized, Latin square sequence of treatments. Cows on the normal-CP diet did not receive infusions.

Infusion treatments on the low-CP diet were 0.9% saline (**SAL**), 1 kg/d of glucose (**GLC**), 1 kg/d of GLC + 75 g/d of BCAA (**GLC+75**), or 1 kg/d of GLC + 150 g/d of BCAA (**GLC+150**). The BCAA were supplied in a 1:1:1 ratio by weight of Leu:Ile:Val. Treatment solutions were mixed daily by completely dissolving GLC and BCAA in 4 L of water set to pH 12.0 with 10 *M* NaOH, adjusting pH to 7.4 with HCl,

Table 1. Ingredient composition and nutrient analysis (% of DM unless otherwise noted) of the 2 experimental diets

Component	Low CP	Normal CP
Ingredient		
Haylage	45.4	28.5
Corn silage	21.7	34.7
Corn, high-moisture	26.7	20.5
Straw, long chop	3.6	3.6
Soy plus[1]	0	4.7
Soybean meal	0	3.0
Canola meal	0	1.5
Fish meal	0	0.5
Biuret	0	0.1
Urea	0.2	0.03
Vitamin/mineral mix[2]	2.4	2.8
Nutrient		
CP	12.6	14.9
Soluble CP, % of CP	32.8	39.1
NDICP,[3] % of CP	23.0	21.7
ADICP,[4] % of CP	10.1	8.4
NDF	35.0	32.8
ADF	21.8	20.4
Lignin	10.5	9.6
NFC	45.9	46.5
Starch, % of NFC	56.6	55.0
Ether extract	3.1	3.1
NE_L,[5] Mcal/kg	1.52	1.59

[1]West Central Cooperative, Ralston, Iowa.

[2]Obtained from Floradale Feed Mill (Floradale, ON, Canada) and consisted of calcium (6.6%), phosphorus (5.0%), sodium (12.5%), sulfur (2.7%), magnesium (6.5%), iron (2,900 mg/kg), zinc (2,833 mg/kg), manganese (2,244 mg/kg), fluorine (483 mg/kg), monensin sodium (460 mg/kg), copper (414 mg/kg), iodine (33.7 mg/kg), cobalt (33.3 mg/kg), selenium (12 mg/kg), vitamin A (436 kIU/kg), vitamin D (70.3 kIU/kg), and vitamin E (1,287 IU/kg).

[3]NDICP = neutral detergent-insoluble CP.

[4]ADICP = acid detergent-insoluble CP.

[5]Calculated according to NRC (2001).

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