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## Abomasal amino acid infusion in postpartum dairy cows: Effect on whole-body, splanchnic, and mammary glucose metabolism

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### ABSTRACT

Nine Holstein cows fitted with rumen cannulas and indwelling catheters in splanchnic blood vessels were used to study the effects of supplementing AA on milk lactose secretion, whole-body rate of appearance (WB-Ra) of glucose, and tissue metabolism of glucose, lactate, glycerol, and  $\beta$ -OH-butyrate (BHBA) in postpartum dairy cows according to a generalized randomized incomplete block design with repeated measures in time. At calving, cows were blocked according to parity (second and third or greater) and were allocated to 2 treatments: abomasal infusion of water ( $n = 4$ ) or abomasal infusion of free AA with casein profile (AA-CN;  $n = 5$ ) in addition to the same basal diet. The AA-CN infusion started with half the maximal dose at 1 d in milk (DIM) and then steadily decreased from 791 to 226 g/d from DIM 2 to 29 to cover the estimated essential AA deficit. On DIM 5, 15, and 29, D[6,6-<sup>2</sup>H<sub>2</sub>]-glucose (23.7 mmol/h) was infused into a jugular vein for 5 h, and 6 blood samples were taken from arterial, portal, hepatic, and mammary sources at 45-min intervals, starting 1 h after the initiation of the D[6,6-<sup>2</sup>H<sub>2</sub>]-glucose infusion. Trans-organ fluxes were calculated as veno-arterial differences times plasma flow (splanchnic: downstream dilution of deacetylated *para*-aminohippurate; mammary: Fick principle using Phe+Tyr). Energy-corrected milk and lactose yields increased on average with AA-CN by 6.4 kg/d and 353 g/d, respectively, with no DIM  $\times$  treatment interaction. Despite increased AA supply and increased demand for lactose secretion with AA-CN, net hepatic release of glucose remained unchanged, but WB-Ra of glucose tended to increase with AA-CN. Portal true flux of glucose increased with AA-CN and represented,

on average, 17% of WB-Ra. Splanchnic true flux of glucose was unaltered by treatments and was numerically equivalent to WB-Ra, averaging 729 and 741 mmol/h, respectively. Mammary glucose utilization increased with AA-CN infusion, averaging 78% of WB-Ra, and increased gradually as lactation advanced. Net portal, hepatic, splanchnic, and mammary fluxes of lactate, glycerol, and BHBA were not affected by AA infusion. Increasing the supply of AA in postpartum dairy cows elevated the WB-Ra of glucose without affecting the true liver glucose release. The greater WB-Ra of glucose with abomasal AA infusion seemed to originate mainly from greater true portal-drained viscera release of glucose. Glucose utilization by the portal-drained viscera was unaffected by abomasal AA infusion, but the exact mechanism behind the greater true portal glucose release could not be assessed in the current study. The increased mammary glucose uptake was in line with the increased milk lactose yield. In early postpartum lactation, the demand for AA seems to be so high that even with increased AA supply, cows have metabolic priorities for AA other than hepatic gluconeogenesis.

**Key words:** dairy cow, transition, glucose, amino acid

### INTRODUCTION

The transition period between late pregnancy and early lactation, including the last 3 wk before parturition to 3 wk after parturition (Grummer, 1995), requires dramatic physiological and metabolic adaptations in the dairy cow. In early lactation, the mammary requirements for energy are several times that of the uterus (Bell, 1995), and nutrient demand to support milk secretion surpasses nutrient intake (Drackley et al., 2001). Consequently, dairy cows enter a period of negative energy balance in the postpartum transition period. To mitigate the deficiency of energy supply, a range of metabolic processes is present in the cow. For example, hepatic gluconeogenesis and mobilization of body reserves are upregulated to compensate the limited energy supply after calving (Ingvarsen, 2006). Despite this known energy deficiency, increasing

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energy supply through glucose infusion immediately after calving has not been successful in increasing milk production (Ørskov et al., 1977; Larsen and Kristensen, 2009). On the other hand, infusing casein during the postpartum period drastically increased milk and milk lactose yields (Ørskov et al., 1977; Larsen et al., 2014).

In cows after peak lactation, casein supplementation increases both whole-body rate of appearance (**WB-Ra**) of glucose and lactose yield (Lapierre et al., 2010). It is not known, however, whether the positive response of milk lactose yield to protein supplementation observed during the close postpartum period (Larsen et al., 2014) is also accompanied by an increased WB-Ra of glucose. In fact, although net portal absorption of AA increased from precalving to early postcalving, net hepatic removal of AA did not change (Doepel et al., 2009; Dalbach et al., 2011), whereas net hepatic release of glucose increased from pre- to postcalving (Doepel et al., 2009; Raun and Kristensen, 2011). In the immediate postpartum period, liver glucose release is supported by important inter-organ transfer of glucogenic carbons, such as lactate from peripheral tissues to the liver, instead of an increased utilization of AA for gluconeogenesis (Larsen and Kristensen, 2013). This suggests that the limited role of AA to hepatic glucose synthesis in the postpartum transition period may be due to their scarcity.

Increased AA supply could also affect portal-drained viscera (**PDV**) metabolism. Infusion of AA could salvage glucose for mammary use by decreasing glucose utilization by the PDV. Both EAA and NEAA contribute significantly to energy expenditure across the PDV (Lobley and Lapierre, 2003). Therefore, with increased AA supply, the PDV could use less glucose either from arterial supply or from starch digested in the small intestine: both mechanisms would salvage glucose for mammary use but only the latter would increase WB-Ra of glucose. It has also been reported that abomasal casein infusion stimulates the secretion of pancreatic  $\alpha$ -amylase (Richards et al., 2003; Swanson et al., 2004), which would increase starch digestion and glucose availability.

Therefore, the hypotheses of the present study were that increasing AA supply postcalving would (1) increase WB-Ra of glucose through an increased net hepatic release, and (2) decrease utilization of glucose by the PDV, both of which would increase glucose availability for mammary usage. Therefore, the objectives of this experiment were to determine how an increased supply of AA during the first 4 wk postpartum affects (1) ECM and lactose yields; (2) WB-Ra of glucose; (3) portal, hepatic, and mammary true fluxes of glucose; and (4) the splanchnic and mammary net fluxes of

lactate and BHBA, and peripheral concentration of NEFA.

## MATERIALS AND METHODS

### *Animals and Treatments*

Four second-lactation, 4 third-lactation, and 1 fourth-lactation Holstein cows were used for the study. Cows were prepared before calving, as described in the companion paper (Larsen et al., 2015), with a rumen cannula of 10 cm diameter and catheters in the portal, 1 hepatic, and 2 mesenteric veins, and 1 mesenteric artery. All cows were offered the same prepartum and postpartum rations (Table 1). The postpartum ration was balanced to provide 100% of energy and MP requirements, for an estimated intake of 25 kg/d and milk production of 40 kg/d, 3.85% fat, and 3.2% CP (NRC, 2001). Prepartum and postpartum, cows were fed diets ad libitum. Postpartum, the daily allowance of TMR was offered in equal meals every even hour from automated feeders (Ankom, Fairport, NY). In addition to TMR, long hay was offered once a day to prevent ruminal disturbances: ad libitum from prepartum through 2 DIM, 2 kg/d (as fed) from 3 to 6 DIM, and 1 kg/d for the remainder of the study. The cows had free access to water and were housed in a tiestall barn. Feed offered and refused was weighed daily. Cows were milked twice daily (0800 and 2000 h) and yield was recorded at each milking. Milk samples were collected for 3 d around each blood sampling day. The experimental protocol was approved by the Institutional Committee for Animal Care of the Sherbrooke Research Centre, and animals were cared for in accordance with the guidelines of the Canadian Council on Animal Care (2009).

Cows were blocked according to parity (second and third or greater) and were assigned to 1 of 2 treatments according to a generalized randomized incomplete block design with repeated measurements in time. The 2 treatments consisted of a continuous abomasal infusion (8.05 kg/d) through a tube installed via the rumen cannula. The treatments were (1) continuous abomasal infusion of water (**CTRL**;  $n = 4$ ) or (2) a mixture of free AA with casein profile (**AA-CN**;  $n = 5$ ), initiated on the day of parturition (DIM 1). The rates of infusion, preparation of the AA solutions, and AA composition of the infusate are detailed in the companion paper (Larsen et al., 2015).

### *Blood Sampling and Labeled Glucose Infusion*

Days of sampling were planned at 5, 15, and 29 DIM. On each of these days, D[6,6-<sup>2</sup>H]-glucose [23.7 mmol/h,

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