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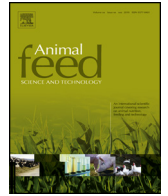


Table 2

Item	Lysine (g/kg)				P-value	Methionine (g/kg)				P-value
	1	2	3	4		1	2	3	4	
DMI (kg)	1.97	1.97	1.97	1.97	0.87	1.97	1.97	1.97	1.97	0.87
Milk yield (kg)	12.2	12.2	12.2	12.2	0.87	12.2	12.2	12.2	12.2	0.87
Milk protein (%)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87
Milk fat (%)	3.2	3.2	3.2	3.2	0.87	3.2	3.2	3.2	3.2	0.87
Milk lactose (%)	4.7	4.7	4.7	4.7	0.87	4.7	4.7	4.7	4.7	0.87
Milk urea N (%)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87
Milk pH	6.5	6.5	6.5	6.5	0.87	6.5	6.5	6.5	6.5	0.87
Milk titratable acidity	1.7	1.7	1.7	1.7	0.87	1.7	1.7	1.7	1.7	0.87
Milk somatic cell count (10 ⁶ cells/ml)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87
Milk yield (kg)	12.2	12.2	12.2	12.2	0.87	12.2	12.2	12.2	12.2	0.87
Milk protein (%)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87
Milk fat (%)	3.2	3.2	3.2	3.2	0.87	3.2	3.2	3.2	3.2	0.87
Milk lactose (%)	4.7	4.7	4.7	4.7	0.87	4.7	4.7	4.7	4.7	0.87
Milk urea N (%)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87
Milk pH	6.5	6.5	6.5	6.5	0.87	6.5	6.5	6.5	6.5	0.87
Milk titratable acidity	1.7	1.7	1.7	1.7	0.87	1.7	1.7	1.7	1.7	0.87
Milk somatic cell count (10 ⁶ cells/ml)	2.7	2.7	2.7	2.7	0.87	2.7	2.7	2.7	2.7	0.87

1, 2, 3 and 4 represent the four experimental groups. Lysine and methionine concentrations in the experimental diets are given in parentheses. Values are means ± SEM. P-values are given in parentheses.

$$\begin{aligned}
 & \text{Lysine (g/kg)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Methionine (g/kg)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{DMI (kg)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk yield (kg)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk protein (\%)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk fat (\%)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk lactose (\%)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk urea N (\%)} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk pH} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk titratable acidity} = \mu + \alpha + \beta + \gamma + \delta \\
 & \text{Milk somatic cell count (10}^6 \text{ cells/ml)} = \mu + \alpha + \beta + \gamma + \delta
 \end{aligned}$$

The model was fitted to the data using the following equation: $Y = \mu + \alpha + \beta + \gamma + \delta$, where Y is the dependent variable, μ is the overall mean, α is the effect of lysine, β is the effect of methionine, γ is the effect of DMI, and δ is the effect of milk production performance. The values of μ , α , β , γ , and δ are given in parentheses.

The values of μ , α , β , γ , and δ are given in parentheses. P-values are given in parentheses.

3. Results

3.1. Dietary amino acids composition, feed intake, and milk production performance

The dietary amino acid composition, feed intake, and milk production performance are given in Table 2. The dietary lysine and methionine concentrations were 2.7 and 3.2 g/kg, respectively. The DMI, milk yield, milk protein, milk fat, milk lactose, milk urea N, milk pH, milk titratable acidity, and milk somatic cell count were 1.97 kg, 12.2 kg, 2.7%, 3.2%, 4.7%, 2.7%, 6.5, 1.7, and 2.7, respectively. The P-values are given in parentheses.

3.2. Estimated digestive flow of essential amino acids

The estimated digestive flow of essential amino acids is given in Table 3. The values are given in parentheses.

Table 3

Least squares means and standard errors for the dependent variables of the 2 × 2 factorial experiment with the 2000 and 2005 wheat straw treatments (2000, 2005) and the 2000 and 2005 wheat straw × 2000 and 2005 wheat straw interaction (2000 × 2005).

Item	Wheat straw				P-value
	2000	2005	2000 × 2005	SE	
DMI (g/kg DM)	1.22	1.25	0.03	0.02	0.10
DMI (g/kg LW ^{0.75})	2.25	2.27	0.02	0.02	0.10
DMI (g/kg LW ^{0.75}) ²	0.22	0.22	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ³	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁴	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁵	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁶	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁷	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁸	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁰	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹¹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹²	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹³	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁴	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁵	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁶	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁷	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁸	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ²⁰	0.27	0.27	0.00	0.02	0.10

Least squares means and standard errors for the dependent variables of the 2 × 2 factorial experiment with the 2000 and 2005 wheat straw treatments (2000, 2005) and the 2000 and 2005 wheat straw × 2000 and 2005 wheat straw interaction (2000 × 2005).

Table 4

Least squares means and standard errors for the dependent variables of the 2 × 2 factorial experiment with the 2000 and 2005 wheat straw treatments (2000, 2005) and the 2000 and 2005 wheat straw × 2000 and 2005 wheat straw interaction (2000 × 2005).

Item	Wheat straw				P-value
	2000	2005	2000 × 2005	SE	
DMI (g/kg DM)	1.22	1.25	0.03	0.02	0.10
DMI (g/kg LW ^{0.75})	2.25	2.27	0.02	0.02	0.10
DMI (g/kg LW ^{0.75}) ²	0.22	0.22	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ³	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁴	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁵	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁶	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁷	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁸	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ⁹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁰	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹¹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹²	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹³	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁴	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁵	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁶	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁷	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁸	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ¹⁹	0.27	0.27	0.00	0.02	0.10
DMI (g/kg LW ^{0.75}) ²⁰	0.27	0.27	0.00	0.02	0.10

Table 5

Item	Lactating				P-value	Non-lactating				P-value
	Control	2.2	22.2	222		Control	2.2	22.2	222	
DMI (kg)	18.2	18.2	18.2	18.2	.22	18.2	18.2	18.2	18.2	.22
Water intake (L)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
Urea N (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22
CP (g)	22.2	22.2	22.2	22.2	.22	22.2	22.2	22.2	22.2	.22

Values are means and standard errors of the mean (SEM) for 10 cows per treatment. P-values are given for the effect of urea concentration on the dependent variables. Lactating and non-lactating cows were pooled for the analysis.

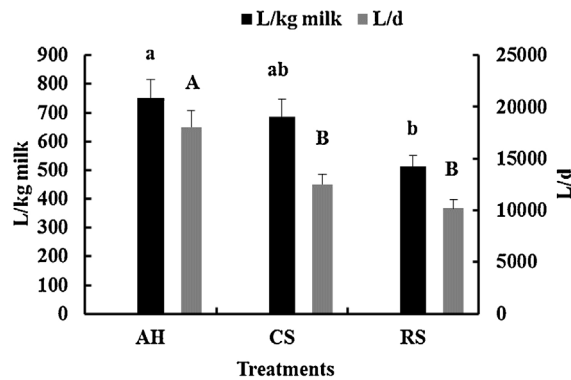


Fig. 1. Milk yield (L/kg milk) and plasma flow (L/d) for three treatments: AH (control), CS (2.2% urea), and RS (22.2% urea). Values are means and standard errors of the mean (SEM) for 10 cows per treatment. P-values are given for the effect of urea concentration on the dependent variables.

(P < 0.05) and plasma flow (L/d) (P < 0.05) were significantly higher in the AH group compared to the CS and RS groups. The effect of urea concentration on milk yield and plasma flow was similar in lactating and non-lactating cows (P > 0.05). The effect of urea concentration on milk yield and plasma flow was similar in lactating and non-lactating cows (P > 0.05).

3.4. Utilization of amino acids by the mammary gland

3.4.1. Mammary plasma flow

Mammary plasma flow (L/d) was significantly higher in the AH group compared to the CS and RS groups (P < 0.05). The effect of urea concentration on mammary plasma flow was similar in lactating and non-lactating cows (P > 0.05).

Table 6

Least squares means (standard errors) for amino acid concentrations in the rumen (g/kg DM) and in the duodenum (g/kg DM) of lactating Holstein cows in the first 12 h of lactation. The values are the means of the two replicates. The values in parentheses are the standard errors. The values in the superscript are the significance of the differences between the two replicates (P < 0.05).

Amino acid	Rumen				P-value	Duodenum				P-value
	Replicate 1	Replicate 2	SEM	SE		Replicate 1	Replicate 2	SEM	SE	
Alanine	1.2	1.2	0.02	0.02	0.98	1.2	1.2	0.02	0.02	0.98
Arginine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Asparagine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Aspartic acid	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Glutamic acid	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Glutamine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Isoleucine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Leucine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Lysine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Methionine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Proline	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Threonine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98
Valine	0.2	0.2	0.01	0.01	0.98	0.2	0.2	0.01	0.01	0.98

Least squares means (standard errors) for amino acid concentrations in the rumen (g/kg DM) and in the duodenum (g/kg DM) of lactating Holstein cows in the first 12 h of lactation. The values are the means of the two replicates. The values in parentheses are the standard errors. The values in the superscript are the significance of the differences between the two replicates (P < 0.05).

3.4.2. Arterial-venous difference and arterial free amino acids supply

The arterial-venous difference in amino acid concentration in the mammary gland was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7). The arterial free amino acid supply was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7).

3.4.3. Clearance rate of amino acids

The clearance rate of amino acids in the mammary gland was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7). The clearance rate of amino acids in the mammary gland was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7).

3.4.4. Mammary uptake of amino acids

The mammary uptake of amino acids was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7). The mammary uptake of amino acids was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7).

3.4.5. Uptake to output ratios for amino acids across the mammary gland

The uptake to output ratios for amino acids across the mammary gland were significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7). The uptake to output ratios for amino acids across the mammary gland were significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7).

4. Discussion

The present study was the first to report the arterial-venous difference in amino acid concentration in the mammary gland of lactating Holstein cows in the first 12 h of lactation. The arterial-venous difference in amino acid concentration in the mammary gland was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7). The arterial free amino acid supply was significantly greater (P < 0.05) for lactating cows in the first 12 h of lactation than for those in the last 12 h of lactation (Table 7).

Acknowledgements

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