# Using Milk Urea Nitrogen to Predict Nitrogen Excretion and Utilization Efficiency in Lactating Dairy Cows[1]

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## ABSTRACT

Because animal agriculture has been identified as a major source of nonpoint N pollution, ways to reduce the excretion of N by production animals must be examined. The objective of this research was to develop and evaluate a mathematical model that integrates milk urea N to predict excretion, intake, and utilization efficiency of N in lactating dairy cows. Three separate digestibility and N balance studies (10 diets, 40 cows, and 70 observations) were used to develop the model, and 19 independent studies (93 diets) were used for evaluation. The driving variables for the model were milk urea N (milligrams per deciliter), milk production (kilograms per day), milk protein (percentage), and dietary crude protein (percentage). For the developmental data set, the model accurately predicted N excretion and efficiency with no significant mean or linear bias for most predictions. Residual analysis revealed that a majority of the unexplained model error was associated with variation among cows. For the independent data set, model prediction error was approximately 15% of mean predictions. A mean of at least 10 cows was determined to be appropriate for model predictions. Target milk urea N concentrations were determined from expected urinary N excretion for cows that were fed according to National Research Council recommendations. Target values calculated in this manner were 10 to 16 mg/dl, depending on milk production. Milk urea N is a simple and noninvasive measurement that can be used to monitor N excretion from lactating dairy cows.

(**Key words**: milk urea nitrogen, nitrogen excretion, modeling)

**Abbreviation key**: **FN** = fecal N excretion, **MUN** = milk urea N, **NI** = N intake, **NUE** = N utilization efficiency, **RMSPE** = root mean square prediction error, **UN** = urinary N excretion.

## INTRODUCTION

Animal agriculture has been identified as a major source of nonpoint N pollution of water resources (45). Animal wastes can contribute to N pollution of the environment as ammonia volatilized to the air, nitrate leached to ground water, and N that runs off to surface water. Improvement of N utilization efficiency (**NUE**) by domestic animals decreases N losses from farms (27). A majority of manure N applied to fields is subsequently lost to water resources even under the best management conditions. Therefore, it is important to reduce manure N output by improving N utilization by the animal. Furthermore, much fertilizer N applied to crops is also lost to water resources before crop uptake. Therefore, the improvement of nutrient utilization by the animal in an effort to reduce the need for crop production further decreases N losses from animal agriculture.

Another concern regarding excess urea in dairy cows is the potential impairment of reproduction because urea, ammonia, or other nitrogenous compounds whose concentrations are associated with urea concentration may be toxic to reproductive tissues (11, 15, 24, 26). As a result, Jordan et al. (24) observed that ova and sperm viability was reduced when dietary protein was excessive. Dairy cows with high blood urea concentrations have been reported to have reduced conception rates (7, 11, 12, 14, 16, 24).

The amount of urea excreted in urine by a cow is directly proportional to the concentration of urea in blood (9), and this amount is proportional to the concentration of urea in milk (41). Therefore, milk urea N (**MUN**) should be a good predictor of urinary N excretion (**UN**) by dairy cows (9, 28).

The objectives of this study were 1) to develop a mathematical model that integrates MUN to estimate UN by lactating dairy cows, 2) to develop this model further to estimate and evaluate additional N nutritional parameters [i.e., fecal N excretion (**FN**), N intake (**NI**), NUE, and DMI], 3) to evaluate the

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TABLE 1. Observational data for model formulation and independent evaluation data sets.

| | Formulation | | | | Evaluation | | | |
|---|---|---|---|---|---|---|---|---|
| Parameter | n | $\overline{X}$ | SD | Range | n | $\overline{X}$ | SD | Range |
| BW, kg | 70 | 598 | 55 | 479–706 | 78 | 592 | 72 | 395–690 |
| DIM | 70 | 132 | 41 | 42–199 | 76 | 108 | 42 | 48–202 |
| Milk, kg/d | 70 | 35.80 | 7.60 | 22.3–50.1 | 89 | 28.38 | 7.13 | 15.1–40.7 |
| Milk fat, % | 70 | 3.30 | 0.44 | 2.34–4.52 | 86 | 4.00 | 0.62 | 2.94–5.69 |
| Milk protein, % | 70 | 3.03 | 0.43 | 2.69–3.54 | 89 | 3.17 | 0.25 | 2.72–3.95 |
| MUN,[1] mg/dl | 70 | 18.46 | 4.73 | 9.7–29.6 | 89 | 15.67 | 4.58 | 1.3–24.7 |
| N Intake, g/d | 70 | 628 | 129 | 400–962 | 89 | 529 | 124 | 214–847 |
| Milk N, g/d | 70 | 176 | 36 | 106–254 | 89 | 144 | 30 | 73–183 |
| Urine N, g/d | 70 | 231 | 74 | 130–411 | 6 | 153 | 79 | 34–214 |
| Fecal N, g/d | 70 | 202 | 29 | 144–276 | 6 | 147 | 32 | 107–180 |
| N Efficiency, % | 70 | 28.30 | 3.72 | 21.1–38.0 | 89 | 27.57 | 4.09 | 18.9–37.0 |
| DMI, kg/d | 70 | 22.4 | 2.8 | 16.1–27.4 | 89 | 19.7 | 3.5 | 10.3–27.7 |
| CP, % of DM | 70 | 17.5 | 1.4 | 13.1–22.4 | 89 | 16.8 | 2.2 | 9.4–23.0 |

[1] Milk urea N.

model with an independent data set, and 4) to establish target ranges for MUN concentrations throughout lactation.

## MATERIALS AND METHODS

### Model Formulation

Three separate digestibility and N balance studies (10 diets, 40 cows, and 70 observations) were used to develop the model (1, 23, 42). Table 1 displays the mean observational data from the formulation data set. Model equations are shown in Table 2.

The renal clearance rate of urea ($C_u$; liters of blood cleared per day) was derived from the following equation (44):

$$C_u = \frac{U_u V}{B_u}$$

where $U_u$ = concentration of urea in urine (grams per liter), V = rate of urine formation (liters per day), and $B_u$ = concentration of urea in blood (milligrams per deciliter). In the regression $U_uV$ = UN (grams per day), $B_u$ = MUN (milligrams per deciliter), and $C_u$ = UN/MUN (100 L/d).

True digestibility of N and metabolic N was estimated using the Lucas test (29) by regressing N disappearance against NI using the following equation (29):

$$D_A = D_t X + m$$

where $D_A$ = N disappearance (grams per day), $D_t$ = true digestibility of N, X = NI (grams per day), and m = metabolic N (grams per day).

Nitrogen intake was predicted from estimated metabolic N and true digestibility of N from the Lucas test, predicted UN, and actual total milk N. Fecal N was predicted from the difference between predicted NI and predicted UN and actual total milk N. The NUE was predicted by dividing actual total milk N by predicted NI and converting the value to a percentage. Dry matter intake was predicted by converting predicted NI to predicted CP intake and dividing by actual dietary CP percentage.

Model predictions were then compared with residual (observed – predicted) values from the data set. Single and multiple regression analyses were performed for residuals against predicted values and other selected variables. Mean bias and linear bias for model predictions were determined (3). Root mean square prediction error (**RMSPE**) was calculated from the following equation (3):

$$\mathrm{RMSPE} = \frac{\sqrt{\Sigma(\text{observed} - \text{predicted})^2}}{\sqrt{n}}.$$

Statistical analyses were performed with JMP$^{\circledR}$ (22), and statistical significance was declared at $P < 0.05$ unless otherwise stated.

### Model Evaluation with Independent Data

Nineteen independent studies (Table 3) were used to evaluate the model. Data from 1 of 19 studies (8) were ultimately excluded from the literature evaluation data set after initial analysis showed that all model predictions for each diet of the study were outside of the 95% confidence interval for each prediction. Table 1 shows the mean observational data from the literature evaluation data set. Single and multi-

TABLE 2. Model equations.

| Prediction[1] | Equation |
|---|---|
| UN,[2] g/d | $12.54 \times$ MUN[3] |
| NI,[4] g/d | (Predicted UN + milk N + 97)/0.83 |
| FN, g/d | Predicted NI − predicted UN − milk N |
| NUE, % | (Milk N × 100)/predicted NI |
| DMI, kg/d | (Predicted NI × 6.25)/dietary CP percentage |

[1] UN = Urinary N excretion, NI = N intake, FN = fecal N excretion, and NUE = N utilization efficiency.

[2] Coefficient for UN prediction obtained from regression of UN versus MUN.

[3] Milk urea N.

[4] Metabolic N and true digestibility coefficient obtained from regression of N utilization versus NI.

ple regression analyses were performed for residuals against predicted values and other selected variables. The RMSPE, mean bias, and linear bias for model predictions were determined (3).

## Target MUN Concentrations

Target MUN concentrations were calculated throughout a standard 305-d lactation from daily urinary N values predicted for cows that were fed diets balanced for CP according to the NRC (35). Driving variables used to calculate UN by lactating dairy cows were milk production (kilograms per day), milk fat (percentage), BW (kilograms), live weight change (kilograms per day), parity (1, 2, or ≥3), and days pregnant. Daily milk production was predicted for a 10,000-kg lactation (49). Changes in milk fat percentage during lactation were predicted (49) at a mean of 3.5%. Lactation curves for daily milk production and milk fat percentage were predicted using the following equation:

$$y_n = an^b e^{cn}$$

where $y_n$ = production character measured at week n of lactation, and a, b, and c = coefficients that define the shape of the lactation curve. Coefficients used for the lactation curves for daily milk production and milk fat percentage were previously published by Wood (49).

Lactation curves for BW and BW change were predicted for a 600-kg cow with a maximum body condition loss of 40 kg during the first 70 DIM. Daily BW was calculated from the following equation (50):

$$C_n = Re^{-kn} + G \exp[h(1 - e^{-g(n-N)})]$$

where $C_n$ = BW at week n of lactation, $Re^{-kn}$ = BW available for loss at week n of lactation, $G \exp[h(1 - e^{-g(n-N)})]$ = total BW gain at week n of lactation, N = weeks of BW loss, and g, h, and k = coefficients that define the shape of the curve. The coefficients that were previously published by Wood (50) were used. Daily BW changes were calculated from the first derivative of the previous equation (50).

To avoid effects of pregnancy on predicted daily urinary N values, pregnancy was assumed to commence at 100 DIM. A sensitivity analysis was performed to determine effects of milk production (kilograms per day), milk fat (percentage), BW (kilograms), and parity (1, 2, or ≥3) on predicted MUN concentrations.

# RESULTS

## Model Formulation

As expected, based on our understanding of physiology (44), when UN was regressed against MUN, the intercept and quadratic terms were not significant. The regression line was UN = 12.54 × MUN (Figure 1; Table 2). The coefficient of the regression represents 1254 L/d of blood completely cleared of urea for excretion in urine, the renal clearance rate of urea. Expressed in terms of BW, the renal blood clearance rate of urea was 1.45 ml/min per kg of BW. Previous research with mature steers (46, 47) showed that the mean renal plasma clearance rates of

TABLE 3. Milk urea N evaluation data set from the literature.

| Experiment | Diets | Cows | Observations |
|---|---|---|---|
| Baker et al. (2) | 4 | 4 | 16 |
| Blauwiekel et al. (4) | 4 | 20 | 70 |
| Broderick (6) | 8 | 40 | 160 |
| Chen et al.[1] (8) | 4 | 36 | 36 |
| Colin-Schoellen et al. (10) | 8 | 28 | 112 |
| Espindola et al. (13) | 3 | 5 | 15 |
| Gonda et al. (18) | 4 | 4 | 16 |
| Huhtanen et al. (21) | 8 | 16 | 16 |
| Mackle et al. (30) | 4 | 32 | 64 |
| Metcalf et al. (31) | 3 | 4 | 12 |
| Metcalf et al. (32) | 4 | 4 | 16 |
| Moorby et al. (33) | 4 | 12 | 48 |
| Oltner and Wiktorsson (37) | 4 | 8 | 24 |
| Pisulewski et al. (38) | 5 | 5 | 25 |
| Robinson et al. (39) | 5 | 25 | 25 |
| Rodriguez et al. (40) | 8 | 24 | 96 |
| Roseler et al. (41) | 5 | 15 | 75 |
| Susmel et al. (43) | 2 | 8 | 8 |
| Wattiaux et al. (48) | 6 | 60 | 60 |

[1] Excluded from final analysis.

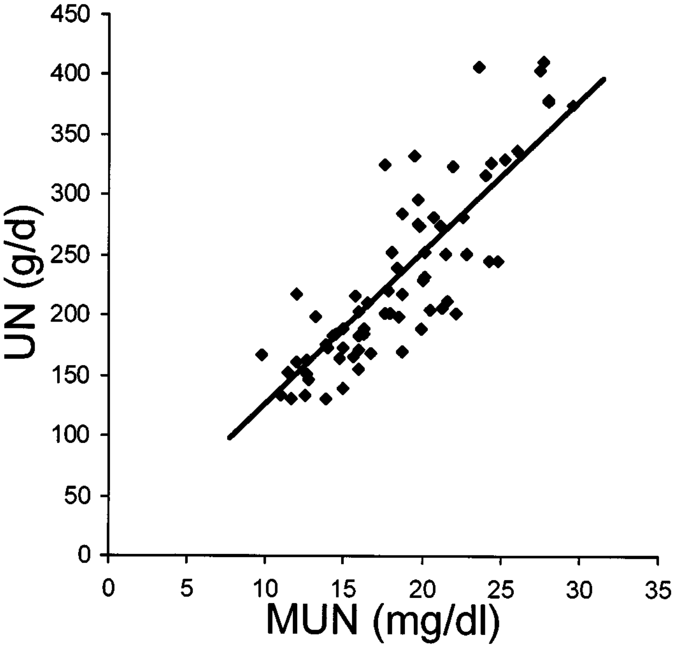

Figure 1. Relationship between milk urea N (MUN; milligrams per deciliter) and urinary N excretion (UN; grams per day); slope = 12.54 ± 0.24.

urea for total UN across treatments ranged from 0.42 to 2.87 ml/min per kg of BW.

The Lucas test (29) calculated metabolic N to be 97 g/d and the true digestibility of N to be 0.83 ($R^2$ = 0.97; Figure 2). In contrast to these results, the NRC (35) predicted metabolic N to be 14.4 g/kg of indigestible DM consumed and predicted RUP digestibility to be 0.80. The present model used a constant amount of metabolic N, regardless of DMI. Metabolic N can result from secretory and excretory materials (29). Results of the Lucas test were used in the prediction of NI (Table 2).

Figure 3 shows the model predictions compared with residual (observed – predicted) values for UN, NI, FN, NUE, and DMI. The RMSPE ranged from 11.0 to 16.9% of mean predictions (Table 4), and mean and linear biases were not significant for most predictions. Regression analyses of residuals versus predictions (Table 4; Figure 3) for the developmental data set did not demonstrate significant mean or linear biases for most parameters. Mean bias was significant for NI and DMI, and linear bias was significant for NUE but accounted for <15% of the RMSPE. Most model error was due to other factors.

Regression analyses of residuals showed that many of the selected variables (Table 5) were significant, but most accounted for only a small proportion of the residual error. Multiple regression analysis revealed >50% of the model error was due to random variation among cows for every prediction except NUE. Body weight or $BW^{0.75}$ variables were significant for all predictions but accounted for <5% of the total model error for any prediction.

## Model Evaluation with Independent Data

The final data set contained data on 89 diets from 18 published studies. Because a total N balance was performed on only 2 experiments with a total of 6 diets, UN and FN predictions were not examined. Figure 4 displays the model predictions compared with residual (observed – predicted) values for NI, NUE, and DMI. The RMSPE ranged from 14.7 to 15.5% of mean prediction (Table 6).

For NI (Figure 4A), the model had no significant mean bias or linear bias (Table 6). Individual regression analysis revealed effects ($P < 0.05$) of all variables except MUN and DIM (Table 7). Multiple regression analysis showed the largest proportion of the RMSPE (81.0 g/d) for NI resulted from variation among experiments.

Together mean bias and linear bias (Table 6) accounted for >30% of the RMSPE of 3.97% for NUE prediction (Figure 4B). The mean bias represented an underprediction of NUE by 0.96%. The negative linear bias indicated an underprediction at a higher

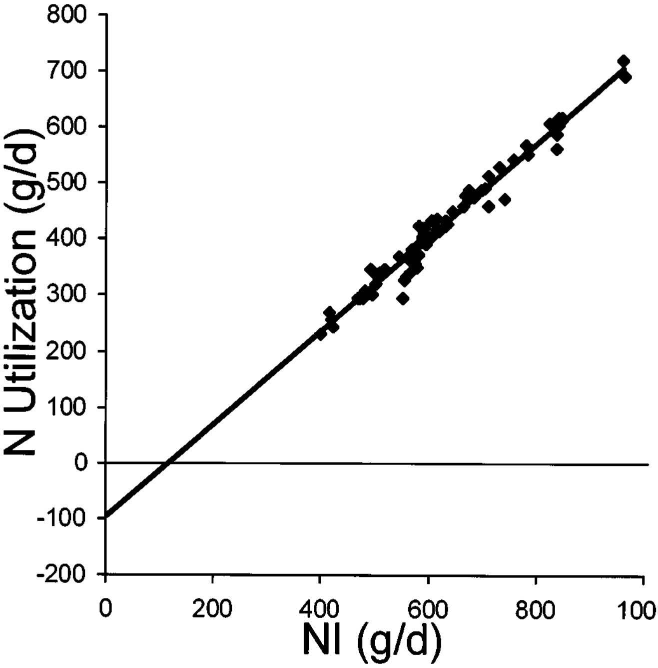

Figure 2. Lucas test used to estimate true N digestibility (0.83 ± 0.02) and metabolic N (97.0 ± 11.7 g/d); $R^2$ = 0.97. NI = N Intake.

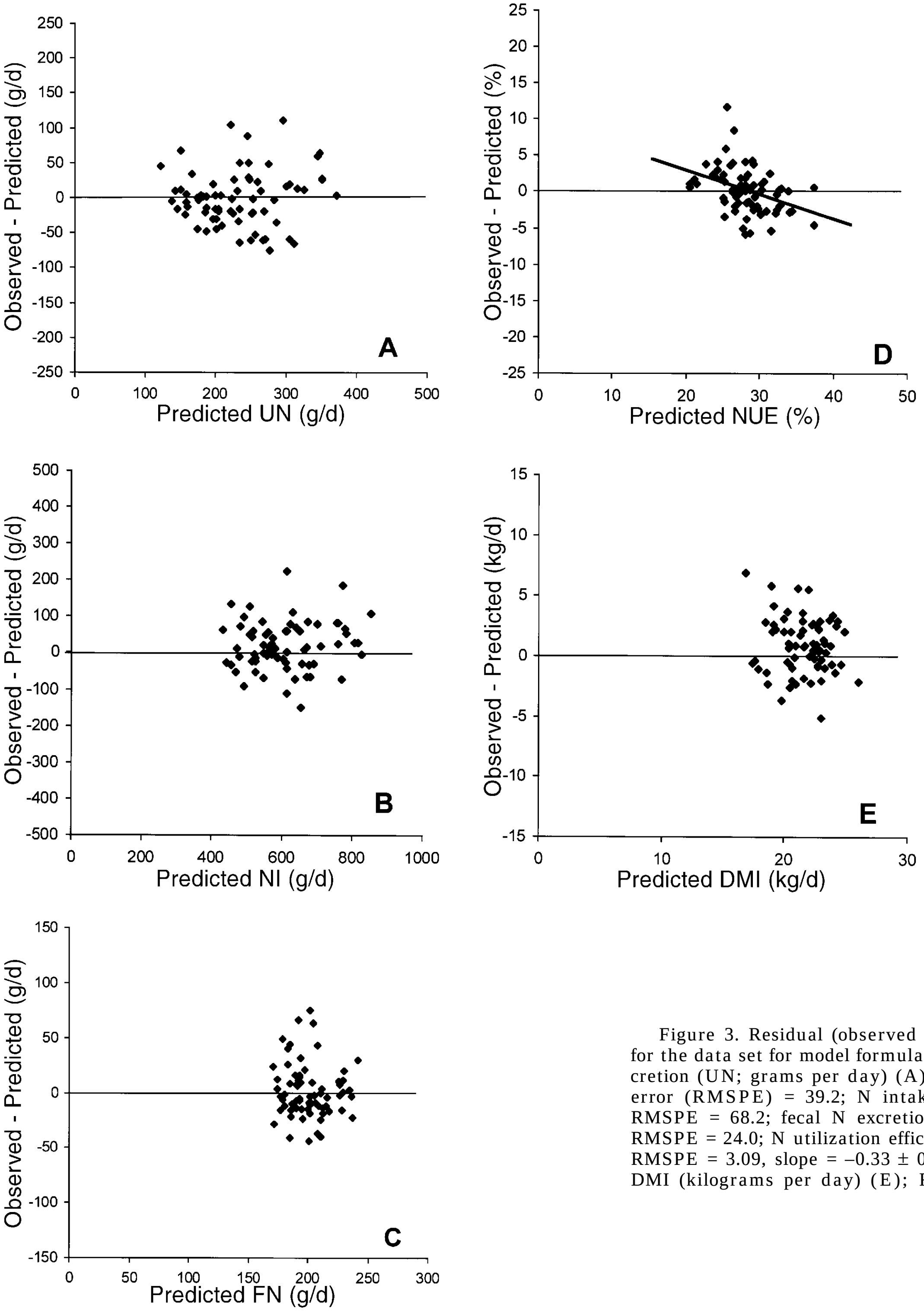

Figure 3. Residual (observed − predicted) values compared for the data set for model formulation with predicted urine N excretion (UN; grams per day) (A), root mean square prediction error (RMSPE) = 39.2; N intake (NI; grams per day) (B), RMSPE = 68.2; fecal N excretion (FN; grams per day) (C), RMSPE = 24.0; N utilization efficiency (NUE; percentage) (D), RMSPE = 3.09, slope = −0.33 ± 0.09, and intercept = 9.61, and DMI (kilograms per day) (E); RMSPE = 2.35.

TABLE 4. Partitioning of root mean square prediction error (RMSPE) for the data set for model developmental.

| Prediction[1] | Mean | RMSPE | CV | Mean bias |  |  | Linear bias |  |  | Residual error[5] |
|---|---|---|---|---|---|---|---|---|---|---|
|  |  |  |  | Bias[2] | $P<$ | Error[3] | Bias[4] | $P<$ | Error |  |
|  |  |  |  |  |  | (%) |  |  | (%) | (%) |
| UN, g/d | 232 | 39.2 | 16.9 | –0.94 | NS[6] | <0.01 | 0.06 | NS | <0.01 | 100.00 |
| NI, g/d | 608 | 68.2 | 11.2 | 19.23 | 0.085 | 7.96 | 0.07 | NS | <0.01 | 92.04 |
| FN, g/d | 200 | 24.0 | 12.0 | 1.26 | NS | <0.01 | –0.09 | NS | <0.01 | 100.00 |
| NUE, % | 28.2 | 3.09 | 11.0 | 0.13 | NS | <0.01 | –0.34 | 0.001 | 14.90 | 85.10 |
| DMI, kg/d | 21.6 | 2.44 | 11.3 | 0.85 | 0.002 | 12.13 | –0.16 | NS | <0.01 | 87.87 |

[1] UN = Urinary N excretion, NI = N intake, FN = fecal N excretion, and NUE = N utilization efficiency.

[2] Mean observation – mean prediction.

[3] Percentage of total prediction error explained by bias.

[4] Slope of residual on prediction.

[5] 100 – [error (percentage) mean bias + error (percentage) linear bias].

[6] $P > 0.10$.

NUE. Individual regression analysis revealed effects ($P < 0.05$) for all selected vari6ables (Table 7). Again, multiple regression analysis showed that the largest proportion of the model error was due to variation among experiments.

The model had no significant mean bias for DMI (Figure 4C) from the independent data set (Table 6). A linear bias ($P < 0.016$) was observed with the model that underpredicted at a higher DMI. Individual regression analysis indicated an effect for all variables of interest (Table 7). As with other predictions from the independent data set, variation among

TABLE 5. Residual analysis for the data set for model formulation (n = 70 cow observations).

| Prediction[2] | $R^2$ | Cow | Diet | Experiment | MUN[1] |  | Milk |  | DIM |  | BW |  | BW$^{0.75}$ |  | CP |  |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
|  |  |  |  |  | Slope[3] | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ |
|  |  | $r^2$ |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Single[4] |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| UN, g/d |  | 0.88*** | 0.45*** | 0.10** | NS[5] | ... | NS | ... | –0.19* | 0.04 | 0.16* | 0.05 | 1.06* | 0.05 | 7.46*** | 0.22 |
| NI, g/d |  | 0.79*** | 0.22* | NS | NS | ... | 1.73* | 0.04 | NS | ... | 0.28* | 0.05 | 1.87** | 0.05 | 6.44** | 0.06 |
| FN, g/d |  | 0.74** | 0.22* | 0.08* | –1.22** | 0.05 | NS | ... | NS | ... | NS | ... | NS | ... | NS | ... |
| NUE, % |  | 0.69* | NS | NS | NS | ... | NS | ... | NS | ... | –0.01* | 0.04 | –0.08* | 0.05 | NS | ... |
| DMI, kg/d |  | 0.77*** | 0.23* | 0.08* | NS | ... | NS | ... | NS | ... | 0.01** | 0.06 | 0.07** | 0.06 | NS | ... |
| Multiple[6] |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| UN, g/d | 0.98 | 0.53[7]*** | 0.37*** | NS | NS | ... | NS | ... | –0.82*** | 0.08 | 39.86*** | 0.04 | –248.00*** | 0.03 | NS | ... |
| NI, g/d | 0.99 | 0.57*** | 0.09*** | 0.11*** | NS | ... | NS | ... | –0.67*** | 0.01 | 44.69** | 0.01 | –272.65** | 0.01 | 45.78*** | 0.04 |
| FN, g/d | 0.97 | 0.75*** | 0.31*** | NS | NS | ... | NS | ... | –0.88** | 0.16 | 26.17*** | 0.01 | –158.43*** | 0.01 | –5.52** | 0.03 |
| NUE, % | 0.43 | NS | NS | 0.24*** | 0.77*** | 0.30 | NS | ... | NS | ... | NS | ... | –0.08** | 0.04 | –1.63*** | 0.31 |
| DMI, kg/d | 0.99 | 0.58*** | 0.08*** | 0.11*** | NS | ... | NS | ... | –0.02*** | 0.01 | 1.27* | 0.01 | –7.71* | 0.01 | –1.40*** | 0.03 |

[1] Milk urea N.

[2] UN = Urinary N excretion, NI = N intake, FN = fecal N excretion, and NUE = N utilization efficiency.

[3] Slope coefficient estimate of variable (predicted – observed) for prediction.

[4] Regression analysis of residuals for UN, NI, FN, NUE, and DMI versus each parameter.

[5] $P > 0.10$.

[6] Multiple regression analysis of residual (predicted – observed) values for UN, NI, FN, NUE, and DMI versus variables, nonsignificant variables were excluded.

[7] Type III partial $r^2$.

*$P < 0.10$.

**$P < 0.05$.

***$P < 0.01$.

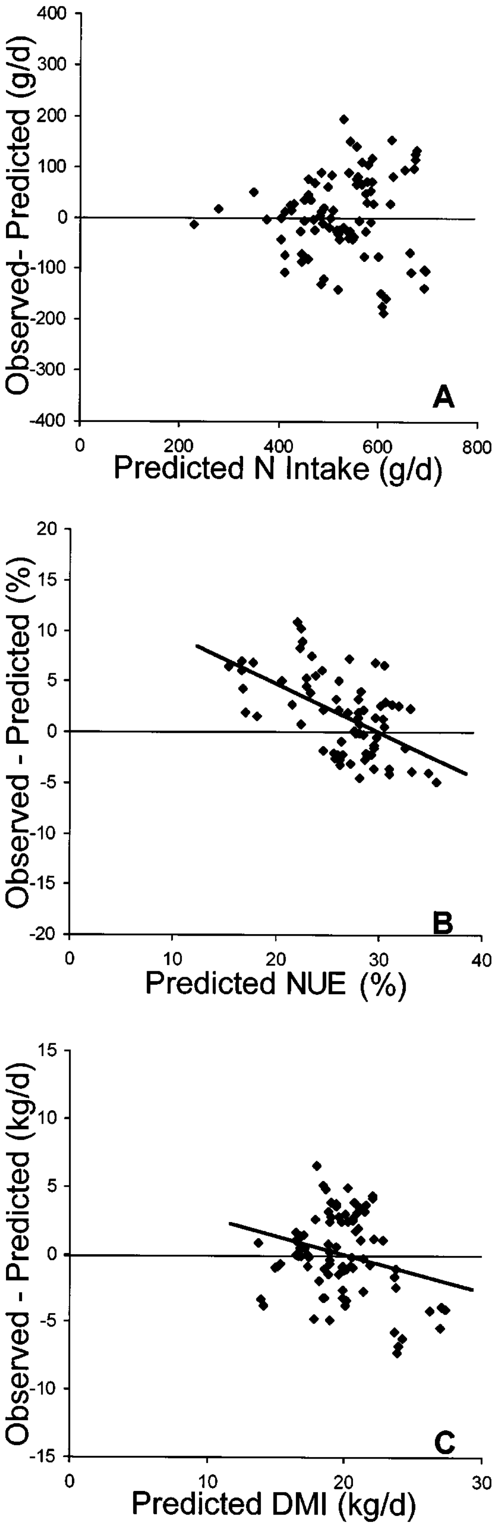

Figure 4. Residual (observed − predicted) values compared for the data set from the literature for independent evaluation with predicted N intake (NI; grams per day) (A), root mean square prediction error (RMSPE) = 81.0; N utilization efficiency (NUE; percentage) (B), RMSPE = 3.97, slope = −0.45 ± 0.08, and intercept = 13.04; and DMI (kilograms per day) (C), RMSPE = 2.96, slope = −0.27 ± 0.11, and intercept = 5.37.

experiments accounted for the highest proportion of model error for the multiple regression analysis.

### Target MUN Concentrations

Figure 5 displays the lactation curve for daily MUN concentrations for a 305-d 8000-, 10,000-, and 12,000-kg lactation predicted from the expected UN when cows were fed according to NRC (35) recommendations. Peak MUN concentrations occurred at 63 DIM, and peak milk production occurred at 35 DIM. The maximum MUN concentration was 15.90 mg/dl with a minimum concentration of 2.62 mg/dl (Table 8). Mean MUN concentration weighted by milk production for the entire 10,000-kg lactation was 13.51 mg/dl.

A 2000-kg increase in milk production per lactation would result in a mean 2.85-mg/dl increase in predicted mean MUN concentration (Table 8). When milk production was decreased by 2000 kg per lactation, mean MUN concentration was reduced to 10.66 mg/dl. A change in milk fat percentage of ±0.5 percentage units changed estimated mean lactational MUN concentration by approximately ±1.70 mg/dl. Each 50-kg increase in BW represents an increase in the mean MUN concentration of 0.42 mg/dl. Changes in parity resulted in an approximate 0.45-mg/dl change in mean MUN concentration. First lactation cows had higher MUN concentrations; mature cows had lower MUN concentrations. In general, an increase in N requirements increased the estimated mean lactational MUN concentration.

## DISCUSSION

### Physiological Basis of Model

The required inputs for the model are MUN, milk N, and dietary CP percentage. This simple mechanistic model uses assumptions from physiology to estimate N excretion, intake, and efficiency by lactating dairy cows. Intake N absorbed into the blood stream of a dairy cow results from the diffusion of ammonia from the rumen and transport of amino acids and peptides from the small intestine. Some of the amino acids and peptides are used for milk production and growth.

Excess amino acids and peptides are deaminated in the liver, and N is converted to urea (44). Ammonia, because it is toxic to the animal, is rapidly converted to urea in the liver (44). The urea enters the circulatory system through the hepatic sinuses, which drain into the hepatic vein (20) and become part of the pool of blood urea N.

TABLE 6. Partitioning of root mean square prediction error (RMSPE) for the data set from the literature for independent evaluation.

| | | | | Mean bias | | | Linear bias | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Prediction[1] | Mean | RMSPE | CV | Bias[2] | $P<$ | Error[3] | Bias[4] | $P<$ | Error | Residual error[5] |
| | | | | | | (%) | | | (%) | (%) |
| NI, g/d | 524 | 81.0 | 15.5 | 2.40 | NS[6] | <0.01 | 0.046 | NS | <0.01 | 100.00 |
| NUE, % | 27.0 | 3.97 | 14.7 | 0.96 | 0.015 | 5.84 | –0.453 | 0.001 | 25.87 | 68.29 |
| DMI, kg/d | 19.5 | 2.96 | 15.2 | –0.01 | NS | <0.01 | –0.273 | 0.016 | 6.59 | 93.41 |

[1] NI = N Intake; NUE = N utilization efficiency.

[2] Mean observation – mean prediction.

[3] Percentage of total prediction error explained by bias.

[4] Slope of residual on prediction.

[5] 100 – [error (percentage) mean bias + error (percentage) linear bias].

[6] $P > 0.10$.

The urea is filtered from the blood by the kidney and is excreted from the body in urine. Blood enters the kidneys through the renal artery (20) and is filtered through the nephrons. This process concentrates the urea for excretion in the urine. Because of counter current flow and differences in membrane permeability in ascending and descending loops of Henle, a concentration gradient for the diffusion of urea into urine is created to remove urea from blood (44). Blood flow through the kidney is constant within an animal, which ensures a constant urea filtration rate (milliliters of blood filtered per minute), regardless of urine volume. With a low volume of urine, urea concentration in the urine would be higher than with a higher volume of urine, but a similar amount of blood would be cleared of urea (44). In addition, with high concentrations of urea in blood, more urea would be removed per minute compared with a low concentration of urea in the blood, but the total amount of blood cleared would remain similar. Therefore, urea excretion is proportional to blood urea concentration.

TABLE 7. Residual analysis for the data set from the literature for independent evaluation (n = 89 treatment observations).

| | | | | MUN[1] | | Milk | | DIM | | BW | | $BW^{0.75}$ | | CP | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Prediction[2] | $R^2$ | Forage[3] | Experiment | Slope[4] | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ | Slope | $r^2$ |
| | | $r^2$ | | | | | | | | | | | | | |
| Single[5] | | | | | | | | | | | | | | | |
| NI, g/d | | 0.46*** | 0.90*** | NS[6] | . . . | 5.76*** | 0.25 | 0.42* | 0.04 | 0.78*** | 0.45 | 4.99*** | 0.44 | 9.03** | 0.06 |
| NUE, % | | 0.39*** | 0.87*** | 0.20** | 0.05 | –0.24*** | 0.18 | –0.02** | 0.07 | –0.04*** | 0.39 | –0.22*** | 0.39 | –0.39** | 0.05 |
| DMI, kg/d | | 0.44*** | 0.91*** | –0.12* | 0.03 | 0.20*** | 0.23 | 0.02** | 0.05 | 0.03*** | 0.41 | 0.17*** | 0.40 | 0.37** | 0.07 |
| Multiple[7] | | | | | | | | | | | | | | | |
| NI, g/d | 0.94 | NS | 0.20[8]*** | –8.61*** | 0.01 | NS | . . . | NS | . . . | 0.41*** | 0.02 | NS | . . . | 15.02*** | 0.01 |
| NUE, % | 0.93 | NS | 0.20*** | 0.56*** | 0.02 | NS | . . . | NS | . . . | NS | . . . | –0.17*** | 0.04 | –0.91*** | 0.01 |
| DMI, kg/d | 0.95 | NS | 0.19*** | –0.35*** | 0.02 | NS | . . . | NS | . . . | NS | . . . | 0.10*** | 0.03 | 0.60*** | 0.01 |

[1] Milk urea N.

[2] NI = N Intake; NUE = N utilization efficiency.

[3] Effect of forage type (alfalfa silage, alfalfa hay, corn silage, grass hay, grass silage, mixed forage, oat silage, or pasture).

[4] Slope coefficient estimate of variable (predicted – observed) for prediction.

[5] Regression analysis of residuals for NI, NUE, and DMI versus each parameter.

[6] $P > 0.10$.

[7] Multiple regression analysis of residual (predicted – observed) values for NI, NUE, and DMI versus variables; nonsignificant variables were excluded.

[8] Type III partial $r^2$.

*$P < 0.10$.

**$P < 0.05$.

***$P < 0.01$.

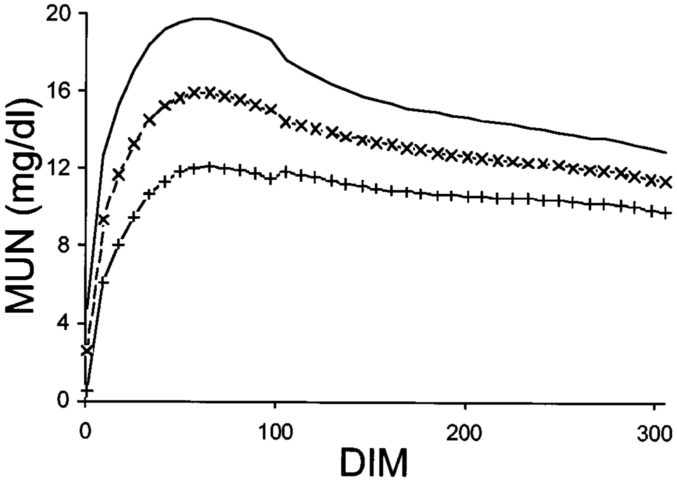

Figure 5. Predicted milk urea N (MUN; milligrams per deciliter) throughout 305-d 12,000-kg (—), 10,000-kg (×), and 8000-kg (+) lactations.

Urea, because it is a small neutral molecule, readily diffuses across cellular membranes. As milk is secreted in the mammary gland, urea diffuses into and out of the mammary gland, equilibrating with urea in the blood. Because of this process, MUN equilibrates with and is proportional to blood urea N (41). This process allows MUN to be an excellent predictor of UN (9, 28).

### Model Function

Predictions from the formulation data set demonstrated that the model would accurately estimate N excretion and efficiency by lactating dairy cows in this data set. The coefficient of variation for predictions was ≤12% for all but the initial UN prediction (Table 4). The mean bias observed for NI and DMI represented an underprediction of only 3% for the model; other predictions showed no mean bias. A linear bias existed for NUE but only accounted for a small proportion of the model error. Overall, mean and linear biases accounted for <15% of the total model error (Table 4). The majority of unexplained model error observed in the multiple regression analysis was due to random variation among cows (Table 5).

Predictions from the independent evaluation data set again demonstrated the accuracy of the model but also highlighted potential problems of overextrapolating the model to data that are outside the range of the original data set. For all three predictions examined, the coefficient of variation was approximately 15% (Table 6), slightly higher than that for the formulation data set. The DMI predictions from the model were less precise than were other DMI predictions (17, 34, 35). The NUE prediction contained both a mean and linear bias, which accounted for approximately 30% of the model error. Still, a large proportion of the model error was unexplained by either a mean or linear bias.

Residual analysis of the literature evaluation data set (Table 7) showed a large effect from experiment to experiment. Cow-to-cow variation may explain a large part of these effects but could not be evaluated because only treatment means were available for analysis. Effects may also be the result of the bias of different laboratory techniques for determining MUN, but, in practice, when the same technique is used for all individuals, this bias would not exist. However, in different experiments for which the same technique was used, an experimental bias could still exist because of time of feeding compared with MUN sampling (19). Although effects of forage type (alfalfa silage, alfalfa hay, corn silage, grass hay, grass silage, mixed forage, oat silage, or pasture) were significant in the individual regression analysis, this variation was completely explained in the multiple regression by differences among experiments. Therefore, forage effects were actually due to differences from experiment to experiment.

Milk production (kilograms per day) exhibited a significant effect on all predictions for the evaluation data set (Table 7) and accounted for 18 to 25% of the unexplained model error in the independent residual analysis. Milk production has been shown previously

TABLE 8. Milk urea N (MUN) estimated from urinary N predictions from the NRC (35).

| | MUN | | |
|---|---|---|---|
| | Mean[1] | Minimum | Maximum |
| Simulation[2] | 13.51 | 2.62 | 15.90 |
| Milk, kg | | | |
| +2000 | 16.36 | 4.79 | 19.76 |
| –2000 | 10.66 | 0.47 | 12.06 |
| Milk fat, % | | | |
| +0.5 | 15.22 | 4.50 | 18.01 |
| –0.5 | 11.81 | 0.74 | 13.80 |
| BW, kg | | | |
| +50 | 13.93 | 2.95 | 16.22 |
| –50 | 13.08 | 2.29 | 15.60 |
| Parity | | | |
| 1 | 13.91 | 2.62 | 16.41 |
| 3 | 12.99 | 2.10 | 15.40 |

[1] Weighted MUN mean for the entire 305-d lactation.

[2] Milk = 10,000 kg per lactation, milk fat = 3.5%, BW = 600 kg, and parity = 2.

TABLE 9. Effect of sample size on 95% CI for cows.

| | Formulation data set[1] | | | | Evaluation data set[2] | | | |
|---|---|---|---|---|---|---|---|---|
| Prediction[3] | 1[4] | 10 | 50 | 100 | 1 | 10 | 50 | 100 |
| UN, g/d | ± 78[5] | ± 25 | ± 11 | ± 8 | NA[6] | NA | NA | NA |
| NI, g/d | ± 136 | ± 43 | ± 19 | ± 14 | ± 162 | ± 51 | ± 23 | ± 16 |
| FN, g/d | ± 48 | ± 15 | ± 7 | ± 5 | NA | NA | NA | NA |
| NUE, % | ± 6.18 | ± 1.95 | ± 0.87 | ± 0.62 | ± 7.94 | ± 2.51 | ± 1.12 | ± 0.79 |
| DMI, kg/d | ± 4.88 | ± 1.54 | ± 0.69 | ± 0.49 | ± 5.92 | ± 1.87 | ± 0.83 | ± 0.59 |

[1] From root mean square prediction error for each prediction from the formulation data set.

[2] From root mean square prediction error for each prediction from the evaluation data set.

[3] UN = Urinary N excretion, NI = N intake, FN = fecal N excretion, and NUE = N utilization efficiency.

[4] Number of cows in sample.

[5] 95% CI = ±2 × root mean square prediction error/$\sqrt{\text{number of cows in sample}}$.

[6] Not applicable.

to affect MUN concentrations (36). Previous research (6, 25, 36, 37) has suggested that the effect of milk production on MUN is caused by the close correlation between production and the protein to energy (ME or $NE_L$) ratio in the diet. In the present study, milk production was not significant during the multiple regression analysis because the model considers milk production and protein content in the prediction of NI. The lack of an effect of milk production demonstrated that the model accurately accounted for this effect. Variation in milk production was caused by differences among experiments, which was completely explained in the multiple regression analysis.

Body weight or $BW^{0.75}$ variables exhibited a significant effect on all predictions for the independent data set (Table 7) for both the individual and multiple regression analyses. Only 3 to 4% of the unexplained model error was attributed to BW variation in the multiple regression analysis. Body weight in the current study had a negative correlation with MUN concentration in lactating dairy cows, which is consistent with previous research (36). On the basis of dilution, when the same amount of urea is formed in the liver, a large animal has more blood than a smaller animal, subsequently causing lower urea concentrations in both blood and milk (36) for the same amount of total urea.

Larger cows are also more likely to have higher renal clearance rates (44) so that mechanistically, inclusion of a BW variable would be appropriate. When the renal clearance rate was derived per kilogram of BW, model precision decreased compared with the use of the same renal clearance rate for all animals regardless of size (data not shown). Variation in BW measurements caused by gastrointestinal fill, milk volume in the udder, and methods of measurement appeared to add so much variation to model predictions that the inclusion of BW in the model was not justifiable. If the model is used for cows that are much smaller than the mean 600 kg in this study, it may be appropriate to adjust the renal clearance rate for BW.

***Sample size.*** As with any method to measure or predict a response, the confidence is greater for the mean as the sample size becomes larger. Table 9 shows the effect of sample size on the 95% confidence interval for each prediction. With the 95% confidence, the UN prediction for an individual observation could vary by ±33% from the actual UN value. However, this confidence interval would be reduced to ±11% for a mean of 10 cows. The current model and analysis can be used to predict variables accurately when averaging 10 or more observations.

Differences among studies in the independent data set demonstrate the need to be cautious about interpretation of model predictions for dairy cows that are not similar to those from the original data set. The original data set included only high producing Holstein cows. The independent data set included high producing Holsteins as well as lower producing Jersey and Red and White Nordic dairy cows. These different cattle breeds could account for differences in the RMSPE for the independent data set compared with the original data set. To establish a 95% confidence interval in model predictions, the RMSPE of the independent data set provided a more conservative estimate.

## Mean Target MUN Concentrations

Expected mean MUN concentrations based on predicted UN by cows that were fed according to NRC

(35) recommendations are most sensitive to differences in milk production. As milk production increases, when cows are fed according to NRC (35) recommendations, MUN concentrations increase because of higher CP intake and N excretion. Although the 50-kg change in BW provided only small differences in estimated MUN concentrations, larger differences in BW can be expected to cause large differences in MUN concentrations.

***Potential research applications.*** A major potential application of this model for research is the prediction of UN from a group of cows. In N balance experiments, the model could use MUN concentrations as a simple noninvasive estimate of UN. Predictions of FN might also be appropriate in N balance studies for a group of cows when treatments do not directly affect digestibility. Model error needs to be considered when making any interpretations from predicted values for an N balance study. An increase in the group size (Table 9) affects the precision of the prediction, and the model should be used with Holstein cows.

A second research application of the model could be estimation of protein intake and DMI from pasture. With measurement of MUN and milk production data, protein intake could be predicted for lactating cows on pasture. With an estimate of the pasture CP content (consumed), DMI could also be predicted. How model predictions would compare with marker or pasture biomass estimates needs further investigation. The current model is best applied to high producing Holstein cows.

***Potential field applications.*** A primary field application of the model is the interpretation of MUN concentrations. When a group of cows has a mean MUN concentration that is higher than the target MUN values at a given level of production (Figure 5), excess protein for the given level of production was probably consumed. A reformulation of the diet at that production level with a lower protein concentration could reduce feed costs. However, before diet reformulation occurs, the specific cause of high MUN should be identified. High MUN can result from a number of nutritional factors, including but not limited to excess protein, inadequate energy, or excess RDP that decreases production and efficiency of N utilization. Close examination of the current diet helps to elucidate the cause of high MUN and leads to appropriate dietary changes to reduce MUN.

A second field application of the model is the estimation of protein intake and DMI from pasture for dairy cows. Current field practices for the estimation of DMI from pasture are labor intensive and require estimates of biomass produced and consumed. A key to the estimation of DMI from pasture would be the accurate characterization of pasture N content. Use of the model for this estimation would require mean input values for a group of cows to predict mean protein intake and DMI.

The model may also be useful for environmental applications. Potential N excretion by dairy cows to the environment could be quantified on a farm or in a watershed. The impacts of changing technologies on dairy farms to reduce N excretion from dairy cows could be monitored through changes in MUN.

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