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Evaluation of Urinary Nitrogen Excretion from Plasma Urea Nitrogen in Dry and Lactating Cows

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ABSTRACT : Data of 42 balance measurements from dry and lactating Holstein cows and blood and urine samples from 24 Japanese Black cows were collected to evaluate the potential for predicting urinary nitrogen (N) excretion from plasma urea nitrogen (PUN). Similar positive correlations were obtained between N intake and apparent N absorption in dry and lactating cows. The regression equations of N intake on urinary N excretion varied in dry and lactating cows, and the difference of urinary N excretion between dry and lactating cows was due to the N secretion into milk. Highly positive correlations were observed between urinary N contents and urinary urea N in Japanese Black cows, and urinary urea N increased with increasing PUN. There were positive correlations between N intake and PUN in dry and lactating cows, but PUN and urinary N excretion in lactating cows were higher than in dry cows. There were positive correlations between PUN and urinary N excretion per BW in dry and lactating cows. Although urinary N excretion could be calculated as (N clearance rate of kidneys)×PUN×BW, high N clearance rate of kidneys, such as 2.08 L/d/kg BW, may be suitable to calculate urinary N excretion in lactating cows, compared with 1.33 L/d/kg BW in dry cows. (**Key Words :** Dry Cow, Lactating Cow, Nitrogen Clearance, Plasma Urea Nitrogen)

INTRODUCTION

Environmental pollution from livestock wastes is a serious problem in Japanese dairy farming, and the dairy industry must develop more efficient milk production, which is in harmony with the environment. The increasing nitrogen (N), phosphorus (P) and potassium (K) excretion in animal wastes is of social concern, and Kojima et al. (2005) reported that decreased intake of N, P and K could decrease the corresponding excretion in dairy cows.

Because N contributes to soil and water pollution as nitrate and to air pollution as ammonia and nitrogen oxide (Tamminga, 1992), it is most important for optimal cow management to decrease N excretion. In practical dairy farming, predicting accurate N excretion rate of cattle is necessary to reduce N excretion, because total collection of feces and urine from cattle is very difficult on a dairy farm. Cattle have adapted to conserving urea nitrogen for recycling to the gut, but excessive dietary protein increases plasma urea nitrogen (PUN) and urinary N excretion in dairy cows (NRC, 2001).

Kohn et al. (2005) reported that PUN or blood urea nitrogen (BUN) was useful as an indicator of protein status and there was a strong linear relationship between BUN and urinary N excretion for cattle. Diets high in protein appeared to stimulate water intake of cows, and then water excess reduced the osmolality of extracellular fluid, resulting in water diuresis and normalization of plasma osmolality (Kume et al., 2008). Renal clearance rate is the rate at which blood is cleared completely of some substances by the kidney, and N clearance rate of kidneys may be a useful index of urinary N excretion. Kohn et al. (2005) estimated that N clearance rates (L of blood cleared of N/d/kg BW) of kidneys in cattle, pigs and rats were 1.3, 3.1 and 4.8, respectively, but the value for cattle included both beef and dairy cattle. Thus, calculating accurate N clearance rates of kidneys in dry and lactating dairy cows is needed to decrease urinary N excretion in practical dairy farming.

In this study, our first objective was to clarify the relationship between PUN and urinary urea N (UUN) in cows fed different amounts of CP. The second objective was

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to evaluate the potential for predicting urinary N excretion from PUN and the N clearance rates of kidneys for dry and lactating cows according to the balance study.

MATERIAL AND METHODS

These experiments were approved by the guide for the care and use of animals in National Agricultural Research Center for Hokkaido Region and Kyoto University Animal Care Committee.

Feeding trial in cows

Data from 24 Japanese Black cows (BW; 444±53 kg) kept at Kyoto University Livestock Farm were collected once a month from August to November 2006. The cows were managed in a paddock. The cows were fed appropriate amounts of wheat bran, Italian ryegrass silage or Sudangrass silage to meet the TDN requirements of breeding cows (Agriculture, Forestry, and Fisheries Research Council Secretariat, 2000), and dietary CP ranged from 12.0 to 16.2%. Blood and urine samples were taken from each cow immediately after feeding small amounts of silage at 06:00 h. At urination between 06:00 to 06:30 h, urine samples were taken in a plastic ladle directly from each cow, and approximate 50 ml of urine was stored at -20°C for chemical analyses. Blood was sampled by jugular venipuncture into heparinized vacuum tubes and centrifuged at 5,000 rpm for 10 min. Plasma urea N and urinary urea N contents were measured by an automatic clinical analyzer (Dri-Chem 3,500 V, Fuji Film, Tokyo, Japan). Urinary N contents were determined as previously described (Kume et al., 2001).

Balance trial in dry and lactating cows

The experiment was conducted as previously described (Kume et al., 2004; Kojima et al., 2005; Kume et al., 2008). Data of 26 and 16 balance measurements were collected from dry and lactating Holstein cows, respectively, in the National Agricultural Research Center for Hokkaido Region. Orchardgrass silage, timothy hay, alfalfa silage and corn silage were offered to meet the TDN requirements of dry cows (Agriculture, Forestry, and Fisheries Research Council Secretariat, 1994), and soybean meal was supplemented for the dry cows fed timothy hay or corn silage because of low CP contents. In 8 lactating cows, orchardgrass silage or alfalfa silage diets were offered in the switch-back trials, and roughage and concentrates were given as a TMR in the ratio of 60:40 to meet the TDN requirements of the cows.

The trials were performed for 14 days in dry and lactating cows. The cows were fed equal amounts of feed at 08:00 and 16:00 h and were given free access to water. Feed refusals and drinking water intake were recorded every day. The lactating cows were milked twice daily, and milk weights were recorded. Cows were housed in individual pens for 7 to 10 days during the feed adjustment period, followed by a 4 to 7 day collection period in a metabolic chamber. The metabolic chamber was maintained at 20°C and 60% relative humidity. Feces and urine from each cow were separately collected daily during the collection period, and composite samples of urine were used for chemical analyses. Nitrogen content in feed, feed refusals, milk, feces and urine were determined as previously described (Kume et al., 2001). Blood was sampled by jugular venipuncture into heparinized vacuum tubes prior to feeding at 08:00 h on the first day of the collection period and PUN was determined by an Automatic analyzer (7250, Hitachi, Tokyo, Japan).

Nitrogen clearance rates of kidney in dairy cows were estimated using the equation of Kohn et al. (2005) in this study. Kohn et al. (2005) reported that there was a strong linear relationship between BUN and urinary N excretion (UNE) for cattle and PUN was assumed to be equivalent to BUN because urea diffuses freely into and out of blood cells. Nitrogen clearance rates (L of blood cleared of N/d/kg BW) of kidneys were calculated as UNE (mg/d)/(BW (kg)× PUN (mg/dl)).

Statistical analyses

The general linear model procedure of SAS (1997) was used to analyze the effect of cows on N balance and plasma composition. Relationships between N intake and N absorption, urinary N excretion or PUN in dairy cows, relationships between UUN and urinary N or PUN in Japanese Black cows and relationships between PUN and urinary N excretion in dairy cows were examined by regression using PROC REG procedure of SAS (1997). Significance was declared at $p<0.05$.

RESULTS

Mean UUN in Japanese Black cows was 676 mg/dl, ranging from 126 to 1,430 mg/dl, and mean PUN was 15.2 mg/dl, ranging from 6.5 to 24.2 mg/dl. There were positive correlations between urinary N content and UUN ($p<0.001$) and between PUN and UUN ($p<0.01$) in cows (Figure 1). The regression equations of urinary N content (X_{UN}) or PUN (X_{PUN}) on UUN (Y_{UUN}) in cows were as follows.

$$Y_{UUN} = 0.941(\pm 0.063)^{***} X_{UN} - 85.6(\pm 55.8)$$
($R^2 = 0.91$, *** $p<0.001$)

$$Y_{UUN} = 44.1(\pm 15.4)^{**} X_{PUN} + 3.3(\pm 243.1)$$
($R^2 = 0.27$, ** $p<0.01$)

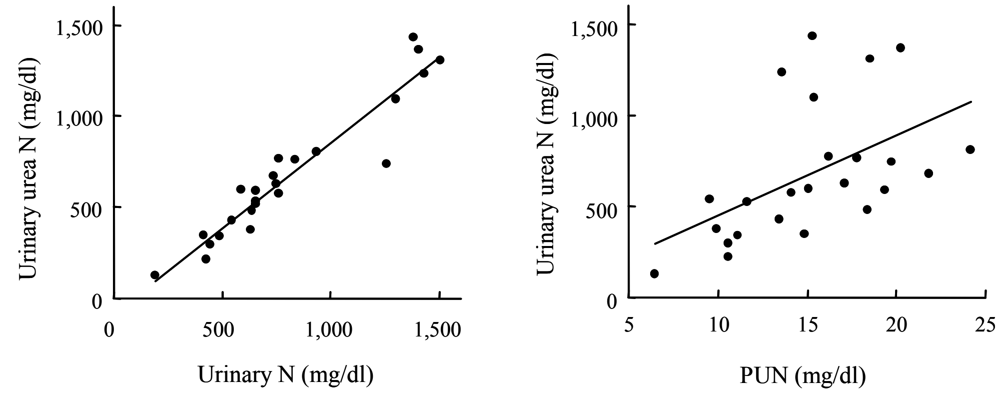

Figure 1. Relationship between urinary N and urinary urea N or relationship between plasma urea N (PUN) and urinary urea N in beef (●) cows.

Milk yield and N secretion into milk in lactating cows was 29.5±3.5 kg/d and 154±17 g/d, respectively. Dry matter intake, water intake, N intake and apparent N absorption of lactating cows were significantly higher ($p<0.001$) than in dry cows (Table 1). There was no significant difference in urinary N content between dry and lactating cows, but PUN, urine volume, urinary N excretion and N clearance rate of kidneys were significantly higher ($p<0.001$) in lactating cows.

There were positive correlations between N intake and apparent N absorption ($p<0.001$), urinary N excretion ($p<0.001$) or PUN ($p<0.01$) in dry and lactating dairy cows and between N intake and urinary N excretion plus N secretion into milk in lactating ($p<0.001$) cows (Figure 2). The regression equations of N intake (X_{Nin}) on apparent N absorption in dry (Y_{Dab}) and lactating (Y_{Lab}) cows, urinary N excretion in dry (Y_{Dex}) and lactating (Y_{Lex}) cows, urinary N excretion plus N secretion into milk in lactating (Y_{Lum}) cows and PUN in dry (Y_{DPUN}) and lactating (Y_{LPUN}) cows were as follows.

Table 1. Urinary and plasma composition of dry and lactating dairy cows (Mean±SD)

Measurement	Cow	
	Dry	Lactating
Number of cows	26	16
BW (kg)	612±70	614±47
DMI (kg/d)	7.5±1.1[b]	20.7±2.2[a]
Water intake[1] (kg/d)	28.0±9.1[b]	98.4±14.9[a]
N intake (g/d)	166±56[b]	557±102[a]
Apparent N absorption (g/d)	110±44[b]	392±77[a]
Urinary N excretion (g/d)	83±40[b]	191±59[a]
Urinary N (%)	0.78±0.23	0.89±0.14
Plasma urea N (mg/dl)	9.9±2.6[b]	14.9±2.9[a]
Urine volume (kg/d)	10.8±4.9[b]	21.9±7.5[a]
N clearance rates[2] (L/d/kg BW)	1.33±0.41[b]	2.08±0.46[a]

[a, b] $p<0.001$.
[1] Drinking water intake plus ingestion of water contained in feed.
[2] Calculated from urinary N excretion/(BW×PUN).

$Y_{Dab} = 0.746(\pm0.048)^{***} X_{Nin}-13.1(\pm8.4)$
($R^2 = 0.91$, *** $p<0.001$)

$Y_{Lab} = 0.723(\pm0.056)^{***} X_{Nin}-10.7(\pm31.4)$
($R^2 = 0.92$, *** $p<0.001$)

$Y_{Dex} = 0.646(\pm0.068)^{***} X_{Nin}-24.4(\pm11.9)$
($R^2 = 0.79$, *** $p<0.001$)

$Y_{Lex} = 0.465(\pm0.092)^{***} X_{Nin}-67.7(\pm52.0)$
($R^2 = 0.65$, *** $p<0.001$)

$Y_{Lum} = 0.582(\pm0.085)^{***} X_{Nin}+21.1(\pm47.9)$
($R^2 = 0.77$, *** $p<0.001$)

$Y_{DPUN} = 0.0269(\pm0.0079)^{**} X_{Nin}+5.4(\pm1.4)^{***}$
($R^2 = 0.32$, ** $p<0.01$, *** $p<0.001$)

$Y_{LPUN} = 0.0182(\pm0.0059)^{**} X_{Nin}+4.8(\pm3.4)$
($R^2 = 0.40$, ** $p<0.01$)

There were positive correlations between PUN and urinary N excretion per kg BW ($p<0.001$) in dry and lactating cows (Figure 2). The regression equations of PUN (X_{PUN}) on urinary N excretion per kg BW (exBW) in dry (Y_{DexBW}) and lactating (Y_{LexBW}) cows were as follows.

$Y_{DexBW} = 0.0154(\pm0.0032)^{***} X_{PUN}-0.020(\pm0.032)$
($R^2 = 0.50$,***$p<0.001$)

$Y_{LexBW} = 0.0274(\pm0.0057)^{***} X_{PUN}-0.094(\pm0.086)$
($R^2 = 0.62$, *** $p<0.001$)

DISCUSSION

Dietary crude protein intake had a large and positive effect on urinary N excretion in dry and lactating cows (Kojima et al., 2005). In this study, apparent N absorption

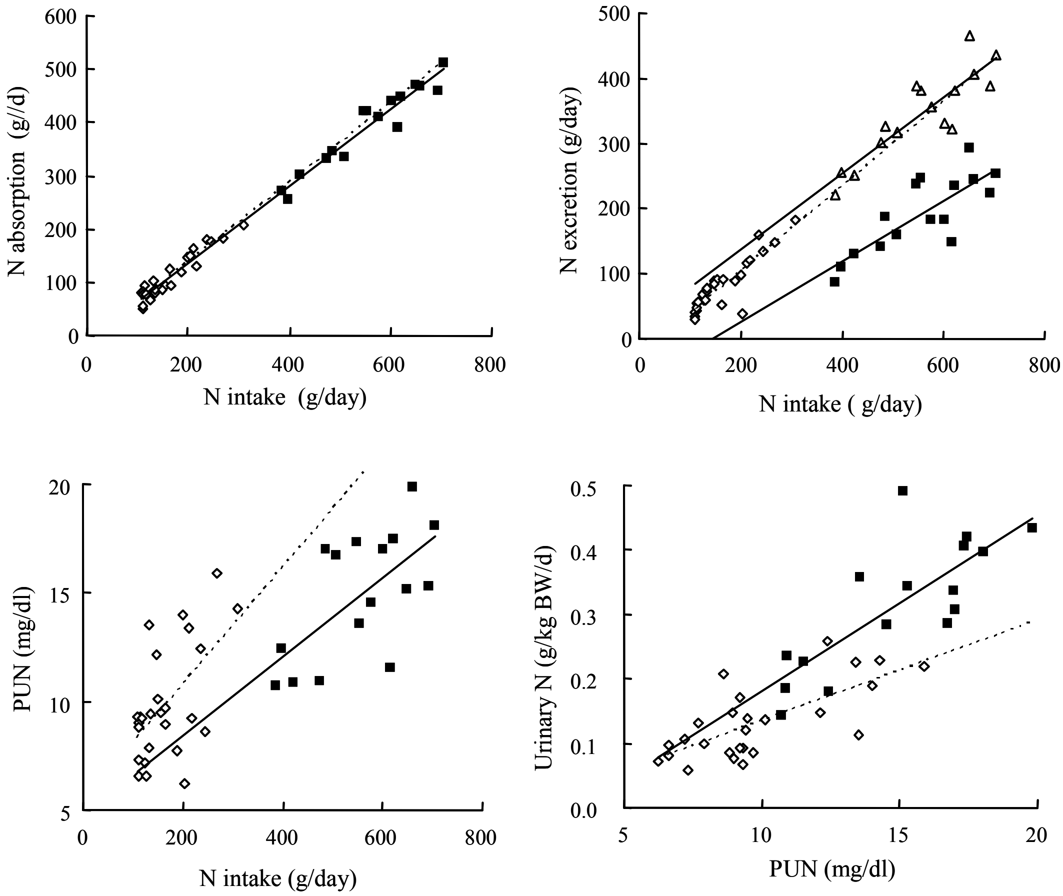

Figure 2. Relationships between N intake and apparent N absorption, urinary N excretion or plasma urea N (PUN) in dry (◊) and lactating (■) cows or relationship between N intake and urinary N excretion plus N secretion into milk (Δ) in lactating cows. Relationships between PUN and urinary N excretion in dry (◊) and lactating (■) cows.

strongly increased as N intake increased in dry and lactating cows. Also, N absorption rates were similar in dry and lactating cows, and approximately 25 to 28% of N intake was excreted in feces. However, the regression equations of N intake on urinary N excretion varied in dry and lactating cows, although urinary N excretion increased as N intake increased. The difference of urinary N excretion between dry and lactating cows was due to the N secretion into milk, because similar regression equations were obtained in N intake on urinary N excretion in dry cows and N secretion into milk plus urinary N excretion in lactating cows. Thus, compared with dry cows, urinary N excretion in lactating cows may be affected by N secretion into milk.

The kidneys regulate N, electrolyte and water excretion to maintain a constant extracellular fluid volume and osmolality. Urinary water excretion was positively related to water availability, amount of water absorbed from the intestinal tract and urinary N and K excretion in cows (Murphy, 1992), and urine volume of cows is determined mainly by the effect of N, K and Na on urine osmolality (Bannink et al., 1999). Because total collection of urine from cattle is very difficult in practical dairy farming, PUN or BUN was useful as an indicator of protein status in cows (Kohn et al., 2005). In this study, highly positive correlations were observed between urinary N content and urinary urea N in cows and 94% of urinary N consisted of urea N, which agreed with data in heifers (Marini and Van Amburgh, 2005). Although PUN above 15mg/dl in cows varied in relation to the urinary urea N, PUN increased as urinary urea N increased.

Excessive dietary protein increased PUN and urinary N excretion in dairy cows (NRC, 2001), and PUN increased exponentially with increasing N content of the diet in heifers (Marini and Van Amburgh, 2005). In this study, PUN in dry and lactating cows increased with increasing N intake, and higher adjusted R^2 were obtained in the prediction models of urinary N excretion per BW from PUN in dry and lactating cows. Based on the fraction of BW because of different size of cows, PUN was useful to estimate urinary N excretion in cows, but the pattern of increasing PUN may be different in dry and lactating cows due to the N secretion into milk.

Urinary N excretion (UNE) could be calculated as: UNE = CR×PUN×BW, where CR represents the calculated N clearance rates of kidneys. In this study, mean N clearance rate of kidneys in dry cows was 1.33 L/d/kg BW, which was similar to the value of 1.3 L/d/kg BW obtained for combined data in dairy and beef cattle (Kohn et al., 2005). However, N clearance rate of kidneys in lactating cows was 2.08 L/d/kg BW. Because N clearance rates of kidneys varied between dry and lactating cows, high N clearance rate of kidneys, such as 2.08 L/d/kg BW, may be more appropriate to calculate urinary N excretion in lactating cows.

These results suggest that urinary N excretion could be calculated from PUN in dry and lactating cows, but an appropriate supply of N to decrease N excretion for dry and lactating cows was not clear. Adachi et al. (2006) reported that a CP level of 14% during a close-up dry period was optimal for primigravid cows according to a N balance study. However, Kamiya et al. (2006) reported that the prepartum diet did not have an apparent effect on N balance in dairy cows after parturition. Dhiman and Satter (1997) suggested that corn silage should constitute one-third to two-thirds of dietary forage dry matter when fed with alfalfa silage to derive maximal benefit and 5 to 15% reduction in the loss of N to the environment. Thus, further study is needed to clarify the optimum supply of N for dairy cows to reduce N excretion and obtain maximal benefit on dairy farms.

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REFERENCES

Adachi, A., T. Kusuhara, I. Nonaka and F. Terada. 2006. Effect of close-up dry period protein level on preparturiental nitrogen balance and lactating performance of primigravid and multiparous Holstein cows. Asian-Aust. J. Anim. Sci. 19:831-836.

Agriculture, Forestry, and Fisheries Research Council Secretariat, 1994. Japanese Feeding Standard for Dairy Cattle (1994). Chuouchikusankai. Tokyo. Japan.

Agriculture, Forestry and Fisheries Research Council Secretariat, 2000. Japanese Feeding Standard for Beef Cattle (2000). Chuouchikusankai. Tokyo. Japan.

Bannink, A., A. Valk and A. M. Van Vuuren. 1999. Intake and excretion of sodium, potassium, and nitrogen and the effects on urine production by lactating dairy cows. J. Dairy Sci. 82:1008-1018.

Dhiman, T. R. and L. D. Satter. 1997. Yield response of dairy cows fed different proportions alfalfa silage and corn silage. J. Dairy Sci. 80:2069-2082.

Kamiya, Y., M. Kamiya and T. Tanaka. 2006. The effect of prepartum diet on nitrogen and major mineral balance of dairy cows during parturition in summer. Asian-Aust. J. Anim. Sci. 19:1415-1421.

Kohn, R. A., M. Dinneen and Russek-Cohen. 2005. Using blood urea nitrogen to predict nitrogen excretion and efficiency of nitrogen utilization in cattle, sheep, goats, horses, pigs, and rats. J. Anim. Sci. 83:79-889.

Kojima, H., S. Kume, K. Nonaka, T. Oshita, T. Kozakai and H. Hirooka. 2005. Effects of feeding and animal performance on nitrogen, phosphorus and potassium excretion by Holstein cows. Anim. Sci. J. 76:139-145.

Kume, S., K. Nonaka, T. Oshita, T. Kozakai and H. Hirooka. 2008. Effects of urinary excretion of nitrogen, potassium and sodium on urine volume in dairy cows. Livest. Sci. 115:28-33.

Kume, S., K. Nonaka, T. Oshita, T. Kozakai and H. Kojima. 2004. Potassium excretion of dry, pregnant and lactating cows fed forage. Nihon Chikusan Gakkaiho 75:179-184 (In Japanese, with English abstract).

Kume, S., T. Toharmat, K. Nonaka, T. Oshita, T. Nakui and J. H. Ternouth. 2001. Relationships between crude protein and mineral concentrations in alfalfa and value of alfalfa silage as a mineral source for periparturient cows. Anim. Feed Sci. Tech. 93:157-168.

Marini, M. R. and M. E. Van Amburgh. 2005. Partition of nitrogen excretion in urine and the feces of Holstein replacement heifers. J. Dairy Sci. 88:1778-1784.

Murphy, M. R. 1992. Water metabolism of dairy cattle. J. Dairy Sci. 75:326-333.

National Research Council. 2001. Nutrient requirements of dairy cattle. 7th. rev. ed. Natl.Acad.Sci., Washington, DC.

Statistical Analysis System (SAS). 1997. SAS/STAT Software: Changes and Enhancement Through Release 6.12. SAS Institute, Cary, NC.

Tamminga, S. 1992. Nutrition management of dairy cows as a contribution to pollution control. J. Dairy Sci. 75:343-357.