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Variations with Breed, Age, Season, Yield, Stage of Lactation and Herd in the Concentration of Urea in Bulk Milk and Individual Cow's Milk

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Carlsson, J., J. Bergström and B. Pehrson: Variations with breed, age, season, yield, stage of lactation and herd in the concentration of urea in bulk milk and in individual cow's milk. Acta vet. scand. 1995, 36, 245-254. – The concentration of urea in the milk of 510 dairy cows in 10 herds was determined at regular intervals for a year. The herds contained approximately equal numbers of Swedish Red and White, and Swedish Holstein cows. The mean ± sd concentration in the samples from individual cows was 5.32 ± 1.13 mmol/l, and the mean concentration in bulk milk was 5.39 ± 0.96 mmol/l. These values indicated that on average the herds were fed too much protein relative to their intake of energy throughout the year

Herd factors had a strong influence on the milk urea concentration. The concentration was lower during the first month of lactation than later in the lactation, and lower when the cows were housed during the winter than when they were grazing. There was a weak positive relationship between the daily milk yield and urea concentration, particularly during late lactation, but there was no relationship with either breed or age. Bulk milk urea was a reliable guide to the average urea concentration of a herd.

profile; nutritional; balance; protein; feeding; dairy production.

Introduction

The concentration of urea in milk is a useful measurement for assessing whether a herd of cows or individual cows are being fed the optimal amounts of protein, whether the ratio of rumen degradable to rumen undegradable protein in the diet is ideal, and whether the balance between the cows' intake of protein and energy is correct *(Carlsson & Pehrson* 1994). Blood urea can be used for the same purpose, because there is a close correlation between the concentrations of urea in blood and milk *(Erbersdobler et al.* 1979, *Oltner & Wiktorsson* 1983, *Refsdal* 1983, *Roseler et al.* 1990), but milk is preferable under most circumstances because it is easier to obtain and is used routinely for other analyses.

To interpret the milk urea concentration correctly it is important to take into account other factors besides the cows' diet. *Carlsson & Bergstrom* (1994) investigated the diurnal variations in concentration and variation during a single milking and between milkings, and the effects of storage and preservation on the concentration of urea in milk samples; they concluded that it was particularly important to take into acount the diurnal variations in milk urea concentration.

There are conflicting reports of the variations in blood or milk urea concentrations during the course of a lactation; *Bruckental et al.* (1980), *Gustafsson et al.* (1987), *Volden et al.* (1992) and *Emanuelson et al.* (1993) all observed lower urea concentrations during the first month of lactation than later, but *Hoffmann & Steinhöfel* (1990) found no differences between the milk urea concentrations at different stages of lactation.

Oltner et al. (1985) and *Gustafsson et al.* (1987) observed a positive correlation between milk yield and milk urea concentration; *Kaufmann* (1982) found a similar relationship but attributed it to the increasing crude protein content of the diet with increasing milk yield rather than to an effect of the milk yield per se. In contrast *Klein* (1984) and *Gustafsson & Carlsson* (1993) found no significant correlations between milk urea and milk yield.

There have been similarly conflicting reports of the effects on milk urea concentration of the age of the cow. An effect of age was observed by *Oltner et al.* (1985) and *Canfield et al.* (1990), but not by *Gustafsson et al.* (1987), *Reinartz* (1988) or *Hoffmann & Steinhöfel* (1990). The influence of breed is also uncertain (*Erbersdobler et al.* 1979, *Wolfschoon-Pombo* 1981).

In a multivariate study *Gustafsson et al.* (1987) found that variation between herds was responsible for 33% of the total variation in milk urea concentration, whereas variation between individual cows was responsible for only 11%. When season, stage of lactation, and yield were included in the statistical model, 63% of the total variation could be accounted for.

Refsdal (1983) suggested that the concentration of urea in bulk milk could be used as an indicator of the nutritional efficiency of a diet being fed to a herd. However, it is necessary to investigate more thoroughly whether the concentration of urea in bulk milk can be relied upon as a measure of the average milk urea concentration of a herd.

In the light of the conflicting results reported above, the present study has been designed to investigate more fully the influence of breed, age, season, milk yield, stage of lactation and herd on the concentration of urea in the milk of individual cows and in bulk milk. It is hoped that the results will make a useful contribution to the evaluation of milk urea concentration as an indicator of the efficiency of practical diets for dairy cows.

Materials and methods

Herds and animals

The investigation was carried out between November 1988 and November 1989 and used 10 dairy herds in south west Sweden. These herds were chosen because they had purebred Swedish Holsteins and Swedish Red and White cows in approximately equal numbers; at least 30% of the cows in each herd belonged to each breed. The herds were all members of the official Swedish Milk Recording System. The mean number of lactating cows in the herds was 37.4, with a range from 29 to 54, and their mean annual milk production was 7,300 kg 4% fat-corrected milk (FCM) with a range from 6,625 to 7,775 kg. Altogether 3,881 individual samples were taken from 510 cows during the study. The median number of samples taken from each cow was 9 with a range from 1 to 12. There were 198 Swedish Holsteins producing a total number of 1,503 samples, 223 Swedish Red and White cows producing 1,696 samples, and 89 crossbred cows producing 682 samples.

All the herds were kept tied up indoors in long stalls from approximately the end of September until the middle of May, and during the rest of the year they were kept on grass and fed supplementary concentrates at milking time. While they were housed they were fed roughage in the form of hay (mean 2.8 kg/day, range 2.0-4.5 kg) and grass silage (mean 6.2 kg dry matter/day, range 2.0-9.0 kg), and concentrates in the form of oats and

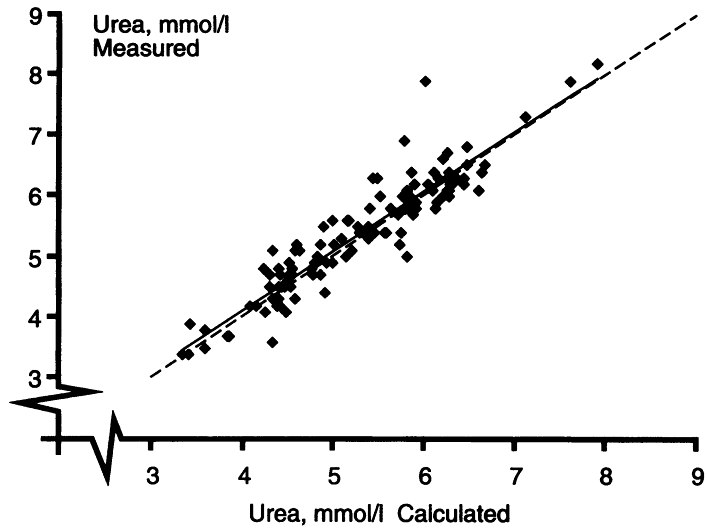

Figure 1: The relationship between the concentrations of urea in bulk milk as measured directly and as calculated. The dotted line is X=Y and the full line represents the calculated regression of Y on X.

barley and a commercial, mineralised protein feed, fed 2 to 4 times a day. The yields of the individual cows were measured once a month, and the amounts of concentrates fed were adjusted to their yields of FCM. The digestible crude protein (DCP) system was used to evaluate the protein fed. The diet fed to 8 of the herds was specified precisely by Milk Recording System advicers, but in the other 2 herds the farmers compounded their own rations, which were reasonably well balanced. During the winter housing period, the rations fed to the cows were evaluated once by a technician; all the feedstuffs were weighed, and the relationship between the metabolizable energy and DCP components of the diets were calculated separately for the low yielding cows (15-20 kg FCM), the middle yielding cows (25-30 kg FCM), and the high yielding cows (>30 kg FCM). All the herds were milked twice a day, starting between 06:00 am and 06:30 am, and between 3:30 pm and 4:30 pm.

At the beginning of the experimental period, 31.1% of the cows were primiparous, and at the end 36.1% were primiparous.

Milk samples

Samples were taken from all the individual cows and from the bulk milk tank once a month, except in July when no samples were taken. The samples from individual cows consisted of a mixture of equal amounts of the milk from the morning and afternoon milkings; samples from the bulk tank were taken no longer than 30 min after the end of the morning milking and after the contents of the tank had been mixed for 5 min. All the samples were preserved with bronopole, kept at 4°C while being transported to the laboratory, and then at –20°C until they were analysed. Samples were missing from 4 bulk milk samplings and from 3 of the monthly samplings of some individual cows involving a total of 89 samples.

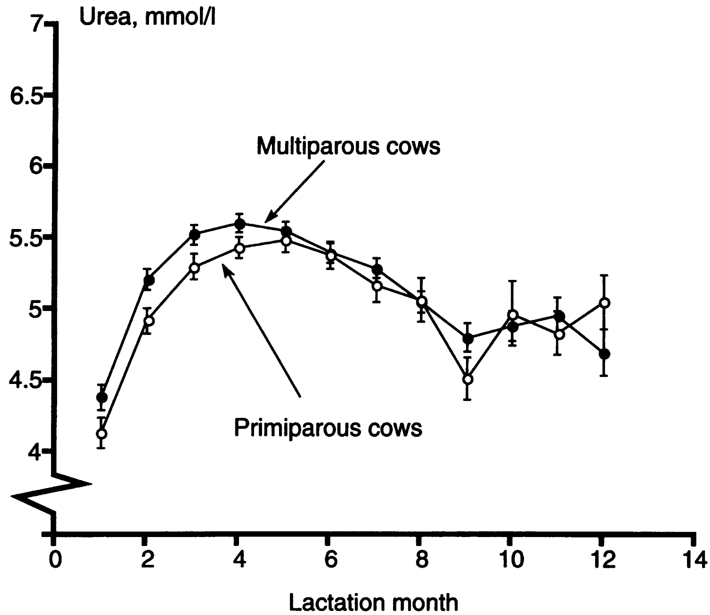

Figure 2: The concentration of urea in milk as a function of the month of lactation during the period when the cows were housed.

Analyses

Milk urea was analysed by the flow injection technique described by *Andersson et al.* (1986). Milk fat content was analysed by an infrared technique at the dairy laboratory.

Statistical analysis

The Statistical Analysis Systems software package *(SAS Institute Inc.* 1987) was used for all the calculations and statistical analyses. To compare the measured concentration of urea in the bulk milk with the bulk milk concentration estimated from the urea concentrations in individual samples, the latter quantity was calculated from the formula:

$$\Sigma Ci * Yi / \Sigma Yi$$

where:

Ci = Urea concentration in the milk of cow i, and

Yi = the milk yield of cow i, uncorrected for fat content

The 2 values were compared by means of the Pearson correlation coefficient.

When comparing the results from different breeds only pure-bred animals were included. The effects of age were investigated by comparing the results from primiparous cows with the results from multiparous cows. The effects of season were investigated by comparing the results obtained while the cows were housed with the results obtained while they were grazing.

The effects of yield were investigated by using 4% fat-corrected milk yields.

Student's T-test (proc T-test) was used for the initial analysis of the effects of breed, age, and season. The effects of breed and age were further analysed for each season, and the effects

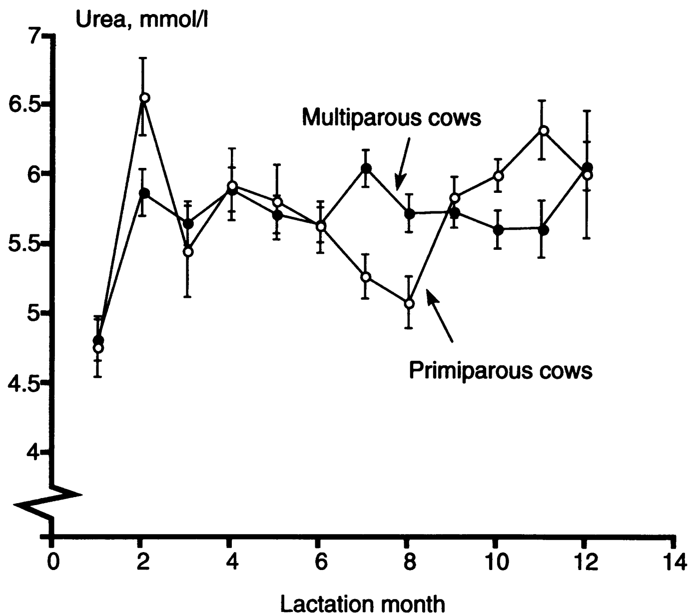

Figure 3: The concentration of urea in milk as a function of the month of lactation during the grazing season.

of breed were also analysed for each age and each season. When groups were not significantly different for a given variable in the initial analysis, the variable was excluded from any further analysis.

For the further analysis of the data proc GLM (general linear models) was used. Stage of lactation was found to have a considerable effect on urea concentrations (Figs. 2 and 3), and 2 stages of lactation were therefore examined in more detail; these were mid-lactation (months 3 to 5) and late lactation (months 8 to 10). Early lactation was excluded because the concentration of urea in milk was very variable during this period, and because the models when applied to the early lactation stages gave no additive information about the high variability except these presented in Figs. 2 and 3. More detailed studies than the present are necessary for explanations of the variability of urea concentration during early lactation – such studies are in progress at our institute. The 2 sets of data used in our study were further reduced by using the mean urea concentration for each cow during each of the 2 seasons. Other factors studied in the GLM models were age, season, yield, and herd.

Thus, in the final analysis, 2 sets of data were used: one set contained the mean urea concentration for each individual cow for those of the months 3 to 5 of a lactation which fell in each season, and the other set contained the corresponding data for months 8 to 10. Some cows provided data for each season in each data set.

Results

The mean ± sd milk urea concentration of the 3,881 individual milk samples was 5.32 ± 1.13 mmol/l (range 1.8-9.1 mmol/l). The mean ± sd daily milk yield was 22.86 ± 6.64 kg FCM (range 2.0-46.0 kg FCM), and the mean ± sd milk fat content was 4.2 ± 0.47% (range 0.4-10.6%). There was a strong correlation between the measured concentration and the calculated concentration of urea in the bulk milk ($r = 0.93$; $p<0.001$) (Fig. 1); the mean ± se; sd measured value was 5.39 ± 0.09; 0.96 mmol/l, compared with 5.28 ± 0.09; 0.92 mmol/l for the calculated value.

The evaluation of the diets fed to the herds revealed that 7 of them were slightly overfed with protein in relation to their intake of energy, 2 received well balanced diets, and 1 was slightly overfed with energy. The mean ratios of MJ/g DCP were 0.92 (range 0.69-1.11) for the low yielding cows, 0.93 (range 0.76-1.09) for the middle yielding cows, and 0.94 (range 0.78-1.09) for the high yielding cows.

There were considerable variations in the concentration of urea in milk with stage of lactation (Figs. 2 and 3). The lowest mean values were consistently observed during the first month of lactation, when they were between 0.8 and 1.9 mmol/l less than a month later, and the difference tended to be larger when the cows were grazing than when they were housed. The shape of the curves was more regular when the cows were housed, and at this time higher urea concentrations were consistently observed at peak lactation than later in lactation. During the grazing season the pattern of urea concentrations in the primiparous cows was different, with an increase during months 9 to 11 of lactation.

No differences between the breeds were observed, either when all the data were considered or when the data were analysed with respect to season, or to age and season together. The effects of breed were therefore excluded from further analysis.

While the cows were housed the multiparous cows had a higher mean urea concentration (5.13 ± 0.04 mmol/l) than the primiparous cows (4.98 ± 0.05 mmol/l), (mean ± se, $p<0.05$), but during the grazing season no difference associated with age was observed.

The mean concentration of urea in milk was significantly lower while the cows were housed (5.16 ± 0.02 mmol/l) than during the grazing season (5.70 ± 0.04 mmol/l) (mean ± se, $p<0.001$).

The results of the GLM analyses are summarised in Table 1. The initial model (model 1) considered the effects due to 4 main variables: yield, age, season, and herd. The model was simplified through models 2, 3, and 4 in which first age, then yield and age, and then herd were eliminated, and finally, in model 5, the data were divided between the grazing and housed seasons and only herd was retained as an explanatory variable. Model 5 retained almost all the explanatory power of model 1. However, yield and season, although not age, also affected milk urea concentration, particularly in late lactation. Yield had a small positive effect on milk urea concentration.

Discussion

In an experimental study of cows fed indoors, *Carlsson & Pehrson* (1994) observed a mean milk urea concentration of 4.74 mmol/l when their diet was balanced in metabolisable energy and DCP. In the present investigation the mean milk urea concentration of the cows when they were housed was 5.16 mmol/l; this higher value was probably due to the fact that the cows were fed too much protein in relation to their energy intake, as was revealed in 7 of the herds by the evaluation of the diets during the winter.

The difference (0.11 mmol/l) between the observed mean concentration of urea in the bulk milk and the lower calculated value was larger than could be explained by the error of the

Table 1: The significance of the relationships revealed by general linear models of the data, considering milk urea concentration as the dependent variable. Mid-lactation is defined as months 3-5 of lactation and late lactation as months 8-10. The yield is the yield of 4% fat corrected milk per cow per day. The seasons are the period when the cows were housed and the period when they were grazing.

Independent variables	Mid lactation		Late lactation	
	p	r^2	p	r^2
Model 1:				
Yield	NS		***[1]	
Age	NS	0.35	NS	0.45
Season	***		***	
Herd	***		***	
Model 2:				
Yield	NS	0.35	**	0 45
Season	***		***	
Herd	***		***	
Model 3:				
Season	***	0.34	***	0.44
Herd	***		***	
Model 4:				
Yield	**	0.04	***	0.16
Age	NS		NS	
Season	*		***	
Model 5:				
Grazing season:				
Herd	***	0.42	***	0.36
Stable season:				
Herd	***	0.46	***	0.55

1 The level of significance p<0.05 is indicated as *, p<0 01 as **, and p<0 001 as ***.

method, the CV of which *Carlsson & Bergström* (1994) estimated to 3%. Since the milk from morning and afternoon milkings differ in urea concentration *(Miettinen & Juvonen* 1990, *Carlsson & Bergström* 1994), one reason for this difference may be that equal volumes of milk were taken out from the afternoon and morning milkings when individual cows were sampled, whereas the proportion of morning milk in a milk tank always is higher than the proportion of afternoon milk. However, the difference was small and the high correlation between the measured and calculated urea concentrations indicates that a measurement of the concentration of urea in bulk milk is a satisfactory measure of the mean concentration of urea in the milk of a herd, as earlier indicated by *Refsdal* (1983). *Wolfschoon-Pombo* (1981) reported that the milk urea concentrations of 2 German breeds of cow were different, although it was accepted that the difference might have been due to the different feeding systems used. *Erbersdobler et al.* (1979) observed no differences in milk urea concentration between breeds, and the present results similarly revealed no difference between the milk urea concentrations of the 2 most common Swedish breeds of cow.

A preliminary analysis suggested that during the winter the multiparous cows had slightly higher milk urea concentrations than the primiparous cows. However, when other factors were taken into account in a general linear model, this difference disappeared, a result in accordance with the results of *Gustafsson et al.* (1987) and *Reinartz* (1988) who also observed no effect of age. However, *Oltner et al.* (1985) did report a difference in milk urea concentration between primiparous and multiparous cows, although their study was based on a small number of animals. Thus, it seems reasonable that milk urea concentrations can be evaluated adequately without considering the age of the cows.

The mean milk urea concentration was significantly higher while the cows were grazing than while they were housed, and a similar difference has been observed previously (*Kaufmann* 1982, *Hoffman & Steinhöfel* 1990, *Carlsson & Pehrson* 1993). The difference is probably due to the high content of easily digestible crude protein in grass relative to its energy content. The application of model 4 (Table 1) indicates that the effect of season was greater in late lactation than in mid-lactation. This effect may have been due to the different strategies for supplementary feeding applied by farmers during the grazing season; in general, grazing is expected to provide a larger fraction of the diet of cows in late lactation, and thus differences between herds would become of less importance.

The general linear models indicated that there was a rather weak positive relationship between milk yield and milk urea concentration, the relationship being clearer during late lactation than during mid-lactation.

The results provide further evidence that differences between herds are the major cause of differences in milk urea concentrations (*Gustafsson et al.* 1987). The important herd factors involved may be the quality of the feedstuffs, the balance between roughages and concentrates fed, the nutritional balances, and the feeding regimens used, in addition to other less important factors. The importance of herd factors strengthens the potential value of measurements of milk urea concentrations in the practical evaluation of dairy rations.

In agreement with several other reports (*Bruckental et al.* 1980, *Gustafsson et al.* 1987, *Volden et al.* 1992, *Emanuelson et al.* 1993) the milk urea concentration was much lower during the first month of lactation than later in lactation, in both age groups and throughout the year. It might have been expected, on account of the difficulty with which a high-yielding cow can satisfy her requirement for energy at this time which leads to breakdown of tissue protein, that the milk urea concentration would have been high, as proposed by *Kaufmann* (1982). Other factors evidently exert effects tending to reduce the urea concentration in the first month of lactation. One such factor may be the inability of cows to ingest sufficient feed early in lactation, which might induce – or be the result of – suboptimal function of the ruminal flora; another factor may be the high risk of metabolic disturbances during the first month of lactation. It is also interesting to note that *Oldham* (1984) discussed the possibility of a nitrogen conserving mechanism in early lactation. It is thus apparent that the reasons for this result certainly require further investigation.

With reference to the low urea concentrations during the first lactation months, misleading information might appear if a great part of the cows in a herd has calved during a short time period, and if bulk milk urea concentrations are measured during the first 1-2 months after that period.

While the herds were housed the mean milk urea concentration decreased during the last 4 to 5 months of lactation. One probable explanation is that in Sweden the central, computerised advisory service recommends feeding a ration with a higher ratio of energy/protein late in lactation than at earlier stages of lactation *(Gustafsson et al.* 1987).

The changes in the concentration of urea in milk were much more regular throughout lactation when the cows were housed than when they were at grass (Figs. 2 and 3), probably because any changes in the nutritional content of the cows' diet were more regular and controlled during the winter. Furthermore, at this time the feed intake of the cows is more likely to be stable and it is easier to feed them according to their requirements. The significant increase in the milk urea concentration of the primiparous cows while they were at grass during the last 3 to 4 months of lactation was probably due to a relative or absolute overconsumption of easily digestible crude protein while their milk yield was declining.

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Sammanfattning

Variation med ras, ålder, årstid, avkastning, laktationsstadium och besättning av koncentrationen av urea i tankmjolk och i mjolk från enskilda kor.

Koncentrationen av urea i mjolk från 510 kor i 10 besättningar bestamdes med regelbundna intervall under 1 års tid. De besattningar som togs med i undersokningen hade ungefär lika många SRB- som SLB-kor. Medelvardet ± sd for samtliga mätningar på individprov var 5.32 ± 1.13 mmol/l och för tankmjolk 5.39 ± 0.96 mmol/l. I medeltal forelåg på besattningsnivå en viss overutfodring med protein i relation till energi under hela laktationen.
Besattningsfaktorer hade starkt inflytande på mjolkureakoncentrationen. Ureavärdena var lagre under forsta laktationsmånaden an senare i laktationen och lagre under stallsasong an under betessasong. Ett svagt positivt samband forelåg mellan dygnsavkastning och mjolkurea, särskilt i slutet av laktationen, medan ureavardena inte i nämnvard grad påverkades av ras och ålder. Tankmjölk befanns vara ett tillforlitligt testmedium for att värdera en besattnings genomsnittliga ureakoncentration.

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