# Monitoring of ammonium pollution from dairy cows farm according of urea content in milk

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Abstract. The objective of this study was to evaluation of urea content in milk to assess the potential of ammonia pollution from farms. Dairy cows in farms were located in different Latvia region with different holding system. Research was conducted under production conditions in four different agricultural holdings located in various places of Latvia and represent different animal housing and feeding technologies. Monthly together with herd control was recorded milk yield and take milk samples was analysed for fat, total protein and lactose (%), urea content (mg dL<sup>-1</sup>) and somatic cell count (SCC). Milk content parameters for total 14,873 milk samples were analysed in accredited milk quality laboratory. The statistical analyses were performed with the SPSS program package. The results acquired show that in three farms (A, B and D) urea content in 59–71% of milk samples comprised 15.1–30.0 mg dL<sup>-1</sup>. However, also in these holdings urea content in 29–41% of samples was below or above the optimum threshold. Unpleasant situation was observed in holding C; there urea content only in 16% of milk samples was within the optimum limits. Calculations of forecasted ammonium pollution emitted daily from agricultural holdings using tie stall housing may comprise 91.4–104.0 g from cow, while amount emitted by freestall holdings using housing may constitute 93.9–95.9 g from cow daily. By using these data, each farm may make calculations and forecast farming efficiency and environmental threats.

Key words: milk urea nitrogen, holding system.

## **INTRODUCTION**

Profitability of dairying industry depends upon successful management of herds: it cannot be organised without suitable monitoring of cows. Monitoring allows organising herd reproduction, arrange correct feeding, improve herd productivity and thus also increase income. The main income in dairying is generated by the milk sold to processing enterprises and is influenced by the milk content, namely composition of fat and proteins; amount and proportions of these components affects milk yield and quality (Verdier-Metz et al., 2001).

Lately interest towards environmental pollution has been growing. In Europe, several regulatory enactments are controlling possible environmental pollution that may arise when performing agricultural activities. In the Netherlands, farms are monitored basing on urea content in milk; that allows specifying possible pollution sources, and notifying farms about preventive actions (Bijgaart, 2003). The optimum amount of urea in milk set in Europe is 15–30 mg dL<sup>-1</sup>.

Data of the National Research Council (in USA) show that nitrogen amount in feed of dairy cows is exceeded on average by 6.6%, thus nitrogen content in urine rises by 16% and by 2.7% in manure. With an aim to calculate amount of nitrogen used, the milk urea content is used, since it is easy to find it out and it does not require collecting and testing special (urine or faeces) samples (Jonker et al., 2002; Broderick & Huhtanen, 2013). Researches show that milk urea characterises content of urea in both blood and urine. Milk urea content reflects losses of crude protein for dairy cow, especially excess in digestive tract, therefore this indicator may be used to assess environmental pollution and digestive efficiency (Broderick & Clayton, 1997; Hof et al., 1997; Burgos et al., 2010).

As compared to other ruminants, dairy cows are able to transform fodder crude protein into milk proteins more effectively and are discharging nitrogen with manure and urine. Nitrogen content in manure may be two, three times higher than one in milk. Thus, as protein volume in feed is increased, not only more milk is produced, bet also threats for environmental pollution are growing. Moreover, costs necessary to prepare fodder rich in proteins are increasing. Share of non-proteins is of a great significance as well, since in European countries and United States of America (USA) it is used to control environmental pollution (Bijgaart, 2003).

The objective of this study was to evaluation of urea content in milk to assess the potential of ammonia pollution from farms.

## **MATERIALS AND METHODS**

Research was conducted:

- Under production conditions and in four different agricultural holdings in Latvia. All holdings are engaged in milk recording program. Holdings included in the research are located in various places of Latvia and represent different animal housing and feeding technologies.
- Data on milk quantities yielded from dairy cows, cow breed, lactation and day in lactation were acquired from monthly herd recording data available in Agricultural Data Centre database.

Within the framework of the research, information on composition of milk samples acquired from the holdings using differing cow housing and dairying facilities was compiled.

Farms taking part in the research are breeding Latvian Brown (LB) and Black and White Holstein (HM) cows, as well as mix of the both breeds (XP). Two large holdings (B and D, 503 and 164 cows, respectively) are keep indoor freestall housing system. Farms recording milk production and uses method A (recording is performed by independent certified person) in line with International Committee of Animal Recording (ICAR) guidelines and legislation of the Republic of Latvia on recording of dairy cows (ICAR, 2011).

In small farms (A and C, 28 and 20 cows, respectively) cows are tie stall housing system, they are not grouped and are grazed in summer. Farms A and C are located in central part of Latvia, near Riga; thus they are limited in availability of agricultural area for high quality grazing areas and meadows, as most of the land is envisaged for construction. Milk production is recording with the help of method B – after acquisition

of certificate, person has a right to perform this task only in own herd. This is recording method suggested by ICAR guidelines.

In all farms cows are milked twice a day. Farms A and C are using milking line. Farm B is milking cows by arranging them into groups, and the process takes place in milking hall with parallel animal placement. All cow groups in farm D are milked in milking hall, animals are placed in herringbone stall.

During the research, milk samples were taken each month in control day. Milk samples were collected from all milking times made during the 24-hour period. During the 26 months, 14,873 milk samples acquired from four farms were analysed. Information on milk samples in breakdown by farm and cow breed has been compiled in the Table 1.

Traits	Farms											
	A (n = 400)		B(n = 10,280)			C (n = 432)		D (n = 3,761)				
Breeds	LB	HM	XP	LB	HM	XP	LB	HM	XP	LB	HM	XP
Number of samples	186	199	15	8,663	1,116	501	393	_	39	389	2,035	1,337
Breakdown of samples, %	47	49	4	84	11	5	91	_	9	10	54	36
Lactation number average	2.30			2.04			3.16			2.20		
Day in lactation, average	185			182			178			185		

Table 1. Analysed milk samples by farm and cow breed

Milk composition was analysed in accredited laboratory for milk quality control.

Research includes evaluation of dairy cow milk productivity traits: yield per cow in control day (yield, kg); content of fat (%), crude protein (%), casein (%), urea (mg dL<sup>-1</sup>) and lactose (%). Indicator used to characterise quality in this research is somatic cell count.

With an aim to research influence left by environmental and selected physiological factors, as well as cow breed on changes in milk composition the multifactor linear model was used; it included the factors fixed:

$$y_{iiklmnsr} = \mu + S_i + Se_i + S_i + L_l + LP_m + Vn + U_s + R_r + e_{iiklmnsr}$$
(1)

where: S – farms (i = 4); Se – season of the year (j = 4); Š – breed (k = 3); L – lactation (l = 1–4); LP – lactation phases (m = 1–6); Vn – health status (n = 6); U – urea content class (s = 4); R – milk yield per cow in control day (r = 6).

Credibility of factors included in linear model of multifactor dispersion analysis was found out at significance level  $\alpha = 0.05$ ; 0.01; 0.001. Influence left by factor was assessed as significant if  $p < \alpha$ . Value of determination coefficient ( $R^2$ ) indicates for how many per cent selected model explains dispersion of the feature researched.

Gradation class average values of the factors researched in model are characterised by last squares mean values (LMS) and standard deviations thereof. The most notable differences among factor gradation class are indicated by various letters: a, b, c etc. if p < 0.05.

With an aim to evaluate and compare research results with other studies and to find out possible nitrogen amount that is wasted by holdings when feeding cows in imbalanced way, content of milk urea that in laboratory was measured as mg dL<sup>-1</sup> was transformed into % (FOSS Analytical, 2005) and afterwards urea volume (g) in control day was calculated in compliance with the guidelines of International Committee for Animal Recording (ICAR, 2011).

Molar mass of urea comprises 60 g moL<sup>-1</sup>, while urea has two nitrogen molecules 28 g moL<sup>-1</sup>. Thus, by calculating proportion from urea content, the content of nitrogen in urea may be found out. By using proportion (28/60) we may recalculate urea content in milk into content of urea nitrogen. In order to be able to compare results with the corresponding data in USA researches and standards, milk urea content was recalculated also into milk urea nitrogen (MUN) content, for further calculations using following formula (Spiekers & Obermaier, 2012):

$$MUN = urea \ content \times 0.46 \tag{2}$$

Also content of urea nitrogen (that in laboratory was measured as per cent urea nitrogen volume (g)) was recalculated in compliance with ICAR guidelines (ICAR, 2011).

$$Volume, kg = (yield, kg \times \% of content)/100$$
(3)

When introducing integrated farming principles, results thereof may be controlled with the help of several indicators. In the research, holdings were assessed by using urea nitrogen volume that is taken from farm together with milk during lactation phase. Recalculation was made per standard lactation (305 days) with the following formula:

## Urea nitrogen volume, kg per cow in lactation = $(MUN volume, kg \times 305)/1,000$ (4)

Possible ammonium pollution in holdings was evaluated with the help of calculations based on model developed in University of California (Burgos et al., 2010):

## Ammonium emission, g per cow daily = $25.0 + 5.03 \times MUN$ content mg dLdL<sup>-1</sup> (5)

Statistical processing of the data was carried out with *MS for SPSS* (SPSS Inc. Chicago, Illinois, USA) and *MS Office* programme *Excel*. Images were created with *MS Office* programme *Excel*.

### **RESULTS AND DISCUSSION**

Farming methods may have significant influence on milk productivity and quality traits. Average milk productivity traits per cow in the control day are shown in the Table 2.

Milk yield in farms participating in research differed significant – it was the lowest in holding C (24.2 kg), while highest in farm D (25.4 kg). Content of crude protein, casein and milk urea varied significantly among the farms. Milk produced in farm B had the highest content of crude protein (3.57%). Moreover, cows in farm B received well-balanced fodder. Casein content varied significant among the holdings; it was the highest in farm B (2.72%) and the lowest in farm A (2.54%).

	Farms							
Traits	A	В	С	D				
	(n = 400)	(n = 10,280)	(n = 432)	(n = 3,761)				
Yield, kg	$25.2\pm0.11^{a}$	$24.9\pm0.05^{b}$	$24.2\pm0.11^{\text{c}}$	$25.4\pm0.05^{a}$				
Crude protein content, %	$3.31\pm0.018^{a}$	$3.57\pm0.008^{b}$	$3.38\pm0.018^{\text{c}}$	$3.53\pm0.008^{d}$				
Casein content, %	$2.54\pm0.014^{a}$	$2.72\pm0.006^{b}$	$2.57\pm0.014^{a}$	$2.69\pm0.006^{\text{c}}$				
Milk urea content, mg dL <sup>-1</sup>	$28.7\pm0.21^{a}$	$29.8\pm0.10^{b}$	$34.1\pm0.22^{\text{c}}$	$30.6\pm0.10^{\text{d}}$				
Fat content, %	$4.25\pm0.045^{a}$	$4.40\pm0.021^{b}$	$4.09\pm0.045^{\text{c}}$	$4.22\pm0.021^{a}$				
Lactose content, %	$4.65\pm0.009^{a}$	$4.71\pm0.005^{b}$	$4.71\pm0.010^{b}$	$4.71\pm0.005^{\text{b}}$				

Table 2. Average cow milk productivity and quality traits in farms studied

 $a_{b,c,d}$  – productivity indicators with unequal letter differed significantly among the farm (p < 0.05).

Urea content in milk also varied among the holdings (from 28.7 mg dL<sup>-1</sup> to 34.0 mg dL<sup>-1</sup>). In farm C feeding was organised on one group, and in summer cows are grazed. Urea content in milk produced in farm C was significant higher (34.0 mg dL<sup>-1</sup>), as compared to other farms. It indicates possible problems in fodder dose balancing and farming. Also Lithuanian scientists (Savickis et al., 2010) emphasize that urea content in milk depends on farm factor.

Milk yielded in farm B had significantly higher fat content (4.40%) and significant lower (4.65%) lactose content. Fat content was the lowest in holding C (4.09%), while lactose content was similar in farms B, C and D (4.71%), moreover it was significantly higher than in farm A.

Urea content in milk shows how correct or suitable is balancing of protein and energy in fodder for cows with various productivity traits. Evaluation of how great is the influence of well-balanced protein and energy amount in fodder dose to milk productivity and quality traits is based on analysis of milk productivity changes depending on urea content thereof (Table 3).

	Urea content, mg d $L^{-1}$							
Traits	3.0-15.0	15.1-30.0	30.1-45.0	45.1 < (n = 470)				
	(n = 1,382)	(n = 8,574)	(n = 4,447)					
Average milk urea content	$13.4\pm0.14$	$23.8\pm0.10$	$35.6\pm0.11$	$50.5\pm0.20$				
in class, mg dL <sup>-1</sup>								
Yield, kg	$24.8\pm0.07^{a}$	$24.8\pm0.05^{a}$	$24.9\pm0.06^{a,b}$	$25.1\pm0.11^{b}$				
Crude protein content, %	$3.41\pm0.012^{a}$	$3.46\pm0.008^{b}$	$3.47\pm0.009^{\text{c}}$	$3.46\pm0.017^{\text{c,b}}$				
Casein content, %	$2.62\pm0.009^{a}$	$2.64\pm0.006^{b}$	$2.65\pm0.007^{b}$	$2.62\pm0.013^{a}$				
Fat content, %	$4.05\pm0.029^{a}$	$4.23\pm0.021^{b}$	$4.36\pm0.023^{\text{c}}$	$4.33\pm0.042^{\text{c}}$				
Lactose content, %	$4.70\pm0.006$	$4.71\pm0.004$	$4.70\pm0.005$	$4.67\pm0.009^{a}$				
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Table 3. Average milk productivity and quality traits depending on urea content

 $a_{b,c}$  – traits with unequal letter differed significantly between the urea level (p < 0.05).

Evaluation of the results acquired shows that all productivity and quality traits researched differed significantly depending on urea content in milk. Milk yield was significantly higher (25.1 kg) if urea content exceeded 45.0 mg dL<sup>-1</sup>. 33% of animals in research had increased urea content in milk. It indicates a problem related to ensuring highly productive dairy cows with fodder dose having adequate proportions of energy and protein (Spohr & Wiesner, 1991; Spann, 1993).

Crude protein content was significant lower (3.41%) in milk yielded from cows the milk urea content of which did not exceed 15.0 mg dL<sup>-1</sup>. The highest crude protein content was observed in milk produced by cows the milk urea of which ranged between 15.1 mg dL<sup>-1</sup> and 30.0 mg dL<sup>-1</sup>. Along with higher milk urea content (above 45.1 mg dL<sup>-1</sup>) crude protein content tends to reduce.

The paper deals with analysing not only influence left by environmental factors on milk productivity and quality traits, but also with studying changes caused by separate physiological factors.

Several scientists (Jonker et al., 2002; Gruber & Poetsch, 2012) emphasize usefulness of crude protein and urea content estimation. Urea content together with crude protein content may be used to assess efficiency of cow feed dose and estimate nitrogen emission into environment.



Analysis of the results found have resulted in average urea content in milk in each farm and breakdown thereof by the content recommended in Europe (Fig. 1).

Figure 1. Milk urea content by farms.

The results acquired show that in three farms (A, B and D) urea content in 59-71% of milk samples comprised  $15.1-30.0 \text{ mg dL}^{-1}$ . However, also in these holdings urea content in 29-41% of samples was below or above the optimum threshold. Unpleasant situation was observed in holding C; there urea content only in 16% of milk samples was within the optimum limits.

The results obtained show how easy urea content in milk allows assessing efficiency with which protein is utilised in each feed dose and identifying potential threats for environmental pollution. Spanish researches on fertilising pastures with slurry, aiming at increasing nitrogen amount in fodder, showed that excessive fertilisation does not give the result desired, since urea content in milk increases, and that, in turn, points to inefficient nitrogen utilisation (Arriaga et al., 2009). Other studies on various fodder doses with 52% and 72% of dry matter and equal protein content (16.5% and 16.4%, respectively) resulted in observation that urea content in milk does not change significantly depending on fodder dose (Agle et al., 2010).

Analysis of the results compiled allows concluded that differences in the results among various holdings arise due to differing feeding methods. Results of this study show differences in average milk urea content among farms, and thus confirm also findings of other researchers about use of milk urea content for effective planning and calculation of effective fodder dose (Jonker et al., 1999). Scientists point out that changes in milk urea content may reflect even 1% change in protein contained by feed dry matter. In the researches mentioned, cows were fed with feed containing 13.0%, 14.0%, 15.0% and 16.0% of protein in dry matter, and significant fluctuations were observed only in urea content, other productivity traits did not change (Zhai et al., 2006).

After evaluation of the research results and correlations, author for successful herd management suggest using milk urea content parameter together with traditional fat and crude protein parameter. Regular control of urea content allows farmer to make sure that fodder doses are effective and ensures that possible problems are discovered and solved in time.

Various researches conducted in Europe have used urea content in milk, while USA studies more often are using other parameter – milk urea nitrogen (MUN) content. In order to be able to compare research results, urea content was recalculated (2) into MUN (Fig. 2) that is used for efficiency control in USA. Advisable MUN content should comprise  $8.0-12.0 \text{ mg dL}^{-1}$  (Kohn et al., 2002; Bucholtz et al., 2007).



Figure 2. Milk urea nitrogen content in researched farms.

Research results show that MUN threshold was exceeded in all farms engaged in the study. Thus it may be concluded that farms have to pay attention to utilisation of protein in fodder and balancing thereof with energy in single feed dose. Findings compiled in the USA regarding fodder protein and MUN content show following: to reach MUN limit 12 mg dL<sup>-1</sup>, it is necessary to reduce protein amount in food to 12.8% in dry matter (Aguilar et al., 2012).

Scientists from countries assessing nitrogen use and efficiency, with which nitrogen in single feed dose is utilised, suggest using urea content parameter to evaluate and plan farming model (Godden et al., 2001; Haig et al., 2002).

Many researchers have proved that milk content traits may be used not only to assess animal productivity, but also to characterise metabolism processes in animal body and thus also to foresee possible illnesses in time and control farming efficiency. Productivity traits characterising body metabolism processes are called biomarkers. Somatic cell count is used to evaluate animal health status, while urea content in milk – to find out protein and energy balance in fodder and assess efficiency of feed protein use, as well as to prognosticate possible risks of metabolism illnesses (ketosis, acidosis) and possible environmental threats. It has been proved that there is significant correlation between milk urea content and nitrogen content in animal urine and manure (Eckersall & Bell, 2010; Burgos et al., 2010; Klein et al., 2011; Spek et al., 2013).

Farm may use urea volume under integrated farming. This indicator points to volume of unused nitrogen that with urine and whey after curd and cheese production gets into waste and afterwards in surrounding environment. By basing calculations (3, 4) on urea and nitrogen amount, volume of nitrogen emitted with milk on average by single cow during lactation may be calculated per each farm (Fig. 3).



Figure 3. Average urea nitrogen emitted with milk by cow in lactation in farms researched.

When calculating average milk urea nitrogen volume that is produced with milk during lactation and possible milk urea nitrogen (MUN) amount at optimum MUN content 8.0 mg dL<sup>-1</sup>, it may be concluded that these indicators differ significantly. Highest urea nitrogen with milk is emitted farm D – 1.116 kg, while at optimum MUN it would be only 0.620 kg, i.e., practically a half less than actual nitrogen volume emitted. Thus each farmer, knowing cost of one protein feed kilogram, may calculate amount of money wasted by farm.

Researches conducted prior have resulted in close positive correlation between milk urea content and milk urea nitrogen; meaning that as urea content in milk increases, also nitrogen content in urine grows, and thus environmental threats and volume of uselessly utilised protein rises as well (Shingfield et al., 2001; Gressley & Armentano, 2007).

As earth population number is growing, issue on food supply becomes increasingly more topical. It is necessary to increase agricultural produce while safeguarding environment. Many researches underline well-balanced agricultural production, seeking for a way to achieve optimum animal productivity with minimum environmental pollution.

S.A.Burgos and other scientists experimented with dairy cows in various lactation days. Cows were fed with fodder doses having various protein contents (15%, 17%, 19% and 21%). Cows received such feeding for six days. In the seventh day, milk, urine and faecal samples were taken, ammonium emission from urine and faecal samples was measured and calculated, and nitrogen content in milk samples was studied. As protein content in fodder dose increased (from 17.2% to 19%), also urine volume of dairy cows grew (from 22.2L daily to 25.6L daily). Basing on the data acquired in the research, calculations were made. Results thereof showed close correlation between ammonium emission with faeces and urine and milk urea content ( $R^2 = 0.85$ ). Basing on the results acquired, scientists worked out equation that is used to control ammonium emission depending on milk urea content (Burgos et al., 2010).

On the basis of this equation (5), possible ammonium pollution in researched farms was calculated (Fig. 4).



Figure 4. Calculated daily ammonium emission per cow in farms.

Calculations show that smallest ammonium pollution would be emitted by holding A, while biggest – by farm C (91.4 g and 104 g, respectively). By using these data, each farm may make calculations and forecast farming efficiency and environmental threats. Netherlands already currently are monitoring and evaluating environmental threats basing on urea content parameter acquired from cow milk monitoring data. Measures taken since 1998, covering monitoring of legislation and farmer control over and correction of fodder protein and energy amount, have produced good result. Already in three years, 12% reduction in ammonium pollution was recorded (Bijgaart, 2003).

Many researchers emphasize that use of urea content is not unambiguous, and it may not be used separately without considering factors influencing changes thereof – not only physiological, but also time, when milk samples were taken, and testing method, as well as laboratory in which testing was performed. Therefore scientists and feeding specialists suggest basing regular herd control on average results calculated that were

obtained from individual animals instead of urea parameter found for total milk produced. If possible, calculations should be made for animals located in the same feeding group (Bijgaart, 2003; Ingvarsten, 2006).

Evaluation of the research results shows that, when planning farming method, it would be useful for each farm not only to consider milk composition, but also to recalculate and assess volume of key milk components in kilograms and grams. Each farmer has to evaluate advantages and disadvantages, and, by using all available milk productivity and quality traits, he/she has to make a decision on the most efficient and environmentally friendly farming method.

### CONCLUSIONS

Milk urea content in holdings using freestall housing where  $29.8-30.6 \text{ mg dL}^{-1}$  comprised in farms using tie stall housing  $28.7-34.1 \text{ mg dL}^{-1}$ , respectively. It was found out that in three farms optimum milk urea content was recorded in 58-70% of milk samples, until in one farm only in 17% of samples.

Milk urea nitrogen (MUN) content in milk produced in the agricultural holdings researched ranged between 13.2 mg dL<sup>-1</sup> and 15.7 mg dL<sup>-1</sup>. MUN volume emitted by single cow during lactation in holdings using freestall housing was higher (from 1.061 kg to 1.116 kg), as compared to farms using tie stall housing (from 1.013 kg to 1.031 kg).

The forecasted ammonium pollution emitted daily from agricultural holdings using tie stall housing may comprise 91.4–104.0 g from cow, withal amount emitted by freestall holdings using may comprise 93.9–95.9 g from cow daily.

Milk productivity traits for farming control should be used together with controlled and known fodder dose, otherwise changes in productivity may not be explained precisely.

All agricultural holdings of milk producing should control urea content in milk yielded from each individual cow on regular basis, while farms engaged in milk monitoring should find out milk urea content for all cows within monthly control.

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