

INNOVATION OF PREDICTION EQUATIONS FOR MILK COMPOSITION ESTIMATION IN MILK RECORDING AT ALTERNATIVE SAMPLING AND HALF A DAY MILKING INTERVAL

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Abstract

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At dairying cost reduction and effort about maintenance of milk recording (MR) range the MR cost decrease is also important. Aim was to revise the older prediction equations for alternative reduced milk sampling (morning – M or evening – E) for assurance of MR reliable results at twice milking a day (TMD) with balanced interval 12/12. Individual milk samples ($n = 619$ animals) were obtained during TMD (M, E and REF; $n = 1\,857$ samples). Fat (F), protein (P), lactose (L) and somatic cell count (SCC) were analysed. Czech Fleckvieh and Holstein breeds were included. The reference values (REF) of day milking were obtained in regular MR (A4P). M and E milking results were related to the REF. Older prediction equations with lower validity because of time and breeding were revised: a) for F from M on REF $y = 0.6932x + 1.381$, where $r = 0.752$ and $P \leq 0.001$; b) for P from M on REF $y = 0.9432x + 0.2274$, where $r = 0.949$ and $P \leq 0.001$; c) for L from E on REF $y = 0.8145x + 0.9095$, where $r = 0.87$ and $P \leq 0.001$; d) for log SCC from E on REF $y = 0.8829x + 0.2367$, where $r = 0.94$ and $P \leq 0.001$. MR cost reduction is possible using method of alternative sampling including corresponding prediction equations.

cow, individual milk sample, fat, protein, somatic cell count

Permanent pressure on cost reduction exists in dairying including milk recording (MR) from economical reasons. Also according to ICAR materials (guidelines, International Committee for Animal Recording, 2011), the MR is still important breeder measure for genetical improvement of dairy cow populations in spite of assertion of modern molecular genetic methods as genomic scanning for advance selection of animals. Information about real own yield of an animal and yield test its offspring is still irreplaceable.

As Hering *et al.* (2005) cite prof. Taufer (1869–1940): *There are no cultivating actions without a yield control and any culture breed dies without genetic transmission control. Therefore the control of parental matter husbandry and nutrient circles cannot only be a temporary*

improvement action but a permanent cultivating work. This fact has to be accepted not only by those managing yield and genetic transmission control but also by those in whose herds the control is carried out.

This is possible to reduce costs on sampling by selection of alternative procedure in months during lactation (morning, evening, morning, evening...) to keep possibility for maintenance of milk recording range. This method A4A (whole day milking in kg and reciprocal sample morning or evening and A4T alternate partial milking in kg of milk and relevant sample) existed always in ICAR materials for MR beside more demanding and more reliable procedure of sampling of all day milkings at milk sample creation by proportional way (A4P) at using of milking weight.

Equations for correction of results of morning and evening milking to composition of whole day milk sample in system of MR in the Czech Republic (CR) for milking twice a day with balanced interval (12/12) are already older regarding changes caused by genetical improvement in dairy cow population during time. These can have more restricted actual validity due to mentioned facts. MR authorized by ICAR organization is very important for confession of international business with breed material. Therefore, used procedures have to be validated. Aim of this work was to carry out a revision and actualization of relevant equations in the CR MR system.

MATERIAL AND METHODS

Animals and their breed, milk samples, conditions of rearing and milking

Cow herds with twice a day milking and regular interval (12/12 hours) were included in observation. There were 2 herds of Czech Fleckvieh (CF) cattle breed in region Plzeň – south (449 dairy cows) and 1 herd with both CF and Holstein (H) breed in region Ústí nad Orlicí (187 dairy cows). Milk recording results of last control year in herds 1, 2 and 3 were: 1) 7056 kg of milk per lactation (305 days), 4.05% of fat, 286 kg of fat, 3.65% of protein and 258 kg of protein; 2) 7519, 3.93, 296, 3.45 and 259; 3) 8310 kg, 3.79%, 315 kg, 3.40% and 283 kg. Milk samples (MSs) were taken stepwise according to localities in period from November 2011 to August 2012. The binding and free stabling were used in stables with pipeline and parlour milking different types from various producers. Dairy cow nutrition was typical for conditions of the Czech Republic in relevant season. Roughage feeding rations were supplemented by concentrates in accordance with feeding standards. The nutrition was characterized by total mixed ration.

Experimental individual MSs were taken by workers of Czech-Moravia Breeders Association (CMBA, organization which is responsible for MR in the CR and member of ICAR) simultaneously with regular MR. MSs were collected in this way that whole day individual milk in MR was represented by one halving sample (A4P) from both milkings (REF) and two samples of partial milking (morning (M) and evening (E)) with interval 12/12 hours ($n = 1\,272$ MSs and 636 animals in total).

Analyses of individual milk samples

MSs were preserved using D&F Control Systems Microtabs (bronopol, 0.03% in milk) tablets and transported under refrigerator conditions ($< 10\text{ }^{\circ}\text{C}$) to laboratory. Samples were analysed in accredited laboratories for milk analyses Buštěhrad and Brno (ZL 1312.2, ZL 1312.3 respectively). These cooperate in routine analytical system of MR (CMBA). MSs were analysed for contents of fat (F; $\text{g}\cdot 100\text{g}^{-1}$), crude protein (P; $\text{g}\cdot 100\text{g}^{-1}$) and lactose (L; lactose

monohydrate; $\text{g}\cdot 100\text{g}^{-1}$) and for somatic cell count (SCC; $10^3\cdot \text{ml}^{-1}$). The instruments MilkoScan FT 6000 (Foss, Denmark) and Bentley 2500 (F, P, L; filter technology of milk infrared spectroscopy; Bentley Instruments, USA) and Somacount 500 (SCC; FC, flow fluor-opto-electronic cytometry; Bentley Instruments, USA) were used for analyses.

Statistical evaluation

Results from recording of partial milkings (M and E) were related to reference (REF) result of MR, it means to whole day milk (HERING *et al.*, 2010; HANUŠ *et al.*, 2011 a, b; CHLÁDEK *et al.*, 2011). Milk indicators such as F, P and L were evaluated in original values. SCCs were evaluated also in logarithmically transformed form because of lognormal frequency distribution occurrence in individual MSs. Programme Excel Microsoft was used for data calculation. Extreme values were excluded using Grubb's test (0.05%) in milk indicator data sets (F, P, L, SCC and log SCC) in M and E groups. This was carried out in whole milking data set (REF) for milk fat as known critical factor of homogeneity and variability of sampling. Number of MSs was eliminated to $n = 619$ animals.

In general, REF value represented the halving sample for whole day milking. The arithmetical means, geometrical means (for SCC), standard deviations and variability coefficients were calculated in sets of components of individual milkings (M and E) and whole day milking (REF). Also median was expressed. The linear and nonlinear (exponential) regression were calculated between M and E milking values (x) and REF (y). Also determination and correlation coefficients or indexes were expressed. The selection of prediction equations for estimation of whole day milk sample composition from M and E milking was carried out. These equations with highest correlation coefficients or indexes were chosen. It means equations with highest measure of REF variability explanation by M or E milking variability.

RESULTS AND DISCUSSION

Basic statistical parameters of observed milk indicators F, P, L and SCC in model files from M, E and whole (REF) milking are shown in Tab. I. F and P mean values were higher in evening ($4.17 > 3.90$ and $3.74 > 3.63\%$) as compared to morning samples. SKÝPALA and CHLÁDEK (2008) found significant difference for milk proteins between E and M samples (3.27 and 3.24%). Further, they showed significantly higher fat content in evening (4.41%) as compared to morning samples (3.95%). It is in accordance with results of this work. Mean values and especially variability characteristics are comparable with other papers on individual MSs (JANŮ *et al.*, 2007 and HANUŠ *et al.*, 2007) of H and CF breed at corresponding milk yield in MR. Only mean protein values were clearly higher in here watched file. That is why this data file has a model

value for determination of prediction equations. Also SCC values, especially low geometrical means (90 and 86 $10^3 \cdot \text{ml}^{-1}$ E and M; Tab. I), showed on relatively good health state of mammary glands of observed dairy cow herds.

The linear dependencies (Tab. II) at all milk indicators were preferred for application. These showed higher prediction reliability in comparison to non linear dependencies (Fig. 1) at majority of indicators also in previous evaluations (HERING *et al.*, 2010; HANUŠ *et al.* 2011 a, b; CHLÁDEK *et al.*, 2011). As

a rule, it was given by higher values of determination coefficient, when linear dependence showed higher percentage of variability explanation of dependent variable value of (REF, whole day milking values) by independent variable value variability (values of M and E alternative sampling; Fig. 2 and 3).

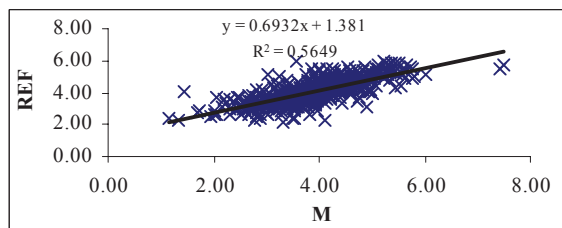
Values of chosen correlation coefficients (Tab. II; at linear relationships) moved from 0.710 ($P \leq 0.001$; F, calculation from E on REF) to 0.949 ($P \leq 0.001$; P, calculation from M on REF). Therefore there were close and very close relationships. There was 50.4%

I: Basic statistical characteristics of milk recording data files, evening (E), morning (M) and reference value (REF, whole day)

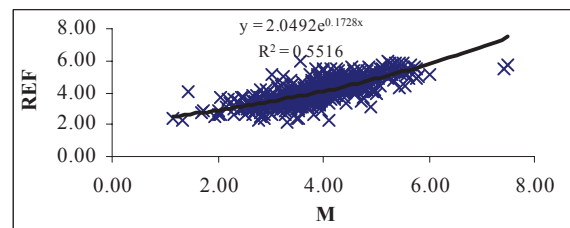
Parameter	Sample	F	P	L	SCC	log SCC
n	E	619	619	619	619	619
x		4.17	3.74	4.83	270.91	1.9557
g						90
sd		0.745	0.412	0.290	751.079	0.605
vx		17.9	11.0	6.0	277.2	
m		4.15	3.73	4.87	95	1.9777
min		1.49	2.74	2.58	5	0.6990
max		6.64	5.14	5.92	9509	3.9781
n	M	619	619	619	619	619
x		3.90	3.63	4.80	258.11	1.9344
g						86
sd		0.777	0.388	0.276	672.224	0.605
vx		20.0	10.7	5.8	260.4	
m		3.88	3.61	4.85	87	1.9395
min		1.14	2.57	2.78	5	0.6990
max		7.49	4.74	5.88	7111	3.8519
n	REF	619	619	619	619	619
x		4.08	3.65	4.84	247.61	1.9634
g						92
sd		0.717	0.385	0.272	601.387	0.568
vx		17.6	10.6	5.6	242.9	
m		4.03	3.64	4.89	85	1.9294
min		2.20	2.69	2.93	5	0.6990
max		5.96	4.75	5.45	6927	3.8405

n = number of cases; x = arithmetical mean; g = geometrical mean; sd = standard deviation; vx = coefficient of variation (%); m = median; min = minimum; max = maximum; F = fat ($\text{g} \cdot 100 \text{g}^{-1}$; %); P = crude protein ($\text{g} \cdot 100 \text{g}^{-1}$; %); L = lactose monohydrate ($\text{g} \cdot 100 \text{g}^{-1}$; %); SCC = somatic cell count ($10^3 \cdot \text{ml}^{-1}$)

F



LIN $y = 0.6932x + 1.381$ $n = 619$
 $R^2 = 0.5649$ $r = 0.752^{***}$



EXP $y = 2.0492e^{0.1728x}$ $n = 619$
 $R^2 = 0.5516$ $r = 0.743^{***}$

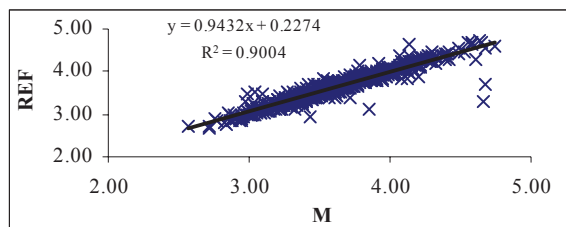
1: Linear and exponential relationship between morning (M) value and REF value (whole milk recording) for fat ($\text{g} \cdot 100 \text{g}^{-1}$; %) in individual milk samples

II: Linear (LIN) and exponential (EXP) equations between alternative milking sample results (E or M) and whole milk recording results (REF, individual milk samples)

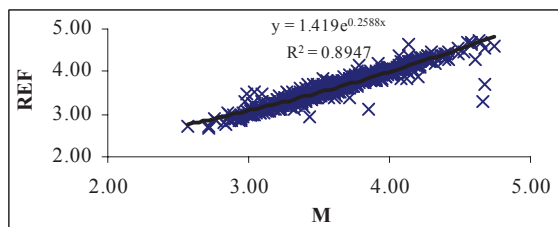
Component	Sample	Relation	Equation	R ²	r	sig
F	M x REF	LIN	$y = 0.6932x + 1.381$	0.5649	0.752	***
		EXP	$y = 2.0492e^{0.1728x}$	0.5516	0.743	***
P		LIN	$y = 0.9432x + 0.2274$	0.9004	0.949	***
		EXP	$y = 1.419e^{0.2588x}$	0.8947	0.946	***
L		LIN	$y = 0.8164x + 0.9252$	0.6875	0.829	***
		EXP	$y = 2.0275e^{0.1811x}$	0.6670	0.817	***
SCC		LIN	$y = 0.8239x + 34.958$	0.8481	0.921	***
		EXP	$y = 68.109e^{0.0012x}$	0.3564	0.597	***
log SCC		LIN	$y = 0.8633x + 0.2935$	0.8465	0.920	***
		EXP	$y = 0.7835e^{0.452x}$	0.7982	0.893	***
F	E x REF	LIN	$y = 0.6833x + 1.2349$	0.5039	0.710	***
		EXP	$y = 1.9848e^{0.1692x}$	0.4858	0.697	***
P		LIN	$y = 0.8847x + 0.3402$	0.8950	0.946	***
		EXP	$y = 1.4617e^{0.2431x}$	0.8919	0.944	***
L		LIN	$y = 0.8145x + 0.9095$	0.7574	0.870	***
		EXP	$y = 1.9391e^{0.1888x}$	0.8922	0.945	***
SCC		LIN	$y = 0.742x + 46.592$	0.8588	0.927	***
		EXP	$y = 69.821e^{0.001x}$	0.3397	0.583	***
log SCC		LIN	$y = 0.8829x + 0.2367$	0.8843	0.940	***
		EXP	$y = 0.7554e^{0.4657x}$	0.8463	0.920	***

R² = determination coefficient; r = correlation coefficient; sig = significance, *** = $P \leq 0.001$

P



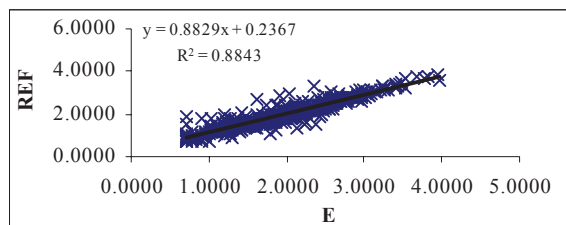
LIN $y = 0.9432x + 0.2274$ $n = 619$
 $R^2 = 0.9004$ $r = 0.949***$



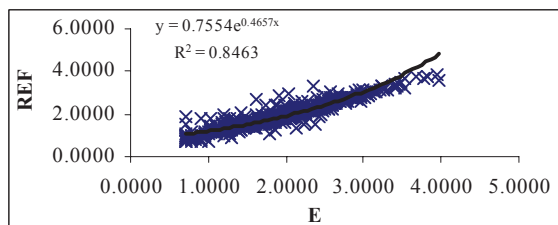
EXP $y = 1.419e^{0.2588x}$ $n = 619$
 $R^2 = 0.8947$ $r = 0.946***$

2: Linear and exponential relationship between morning (M) value and REF value (whole milk recording) for crude protein (g.100g⁻¹; %) in individual milk samples

log SCC



LIN $y = 0.8829x + 0.2367$ $n = 619$
 $R^2 = 0.8843$ $r = 0.940***$



EXP $y = 0.7554e^{0.4657x}$ $n = 619$
 $R^2 = 0.8463$ $r = 0.920***$

3: Linear and exponential relationship between evening (E) value and REF value (whole milk recording) for log SCC (somatic cell count, 10³.ml⁻¹) in individual milk samples

in F case and in P case as many as 90.0% of variability in REF values explainable by variability in values of partial samples which were taken by alternative variants (M and E). Also the REF prediction is least confident in F values. These results are in accordance with previous evaluation (HERING *et al.*, 2010). KLOPČIČ *et al.*, (2003) found higher prediction reliability for P estimation as compared to F, which is similar finding as in this paper (Tab. II). In SCC and L the predicted values then have middle rate of probability. At linear estimation of REF SCC in original units (Tab. II) from alternative values (M and E) was the reliability (coefficients of determination) comparable with log SCC. In theory, the estimation from log SCC values should be more advantageous but real values did not confirm it.

These results completed and extended the pieces of knowledge about estimations of resulting total values of MR from various methodical variants and modifications of individual milk sampling. Further, these results innovated the older used equations. The results contributed to line of published papers (GILBERT *et al.*, 1972; LEE and WARDORP, 1984; HARGROVE, 1994; PALMER *et al.*, 1994; CASSANDRO *et al.*, 1995; LEE *et al.*, 1995; JOVANOVAČ *et al.*, 2005; ROELOFS *et al.*, 2007; GANTNER *et al.*, 2008, 2009; JENKO *et al.*, 2010).

Regarding relatively high tightness of dependencies (reliability) of obtained prediction equations for observed milk indicators this is possible to state hypothetically that pertinent possible differentiation of data set and its evaluation

for instance according to breeds, (eventually animal blood proportions in individual cases), lactation number or stage probably would not bring any essential increase of estimation reliability. Also this evaluation came out right of real conditions of practical application.

CONCLUSION

The MR cost reduction at achievement of relatively reliable results for genetic improvement in dairy cow population is possible due to use of alternative milk sampling (morning and evening) of individual animals in MR during lactation and updated prediction equations for assesment of values of whole day milk composition in the system of twice a day milking with balanced interval (12/12 hours). The milk yield and composition results in milk recording are used, beside practical purposes, also for scientific evaluation of lactation relationships (e.g. KOPEC *et al.*, 2011; GOLEBIEWSKI *et al.*, 2011; ŘEHÁK *et al.*, 2012). Recommended prediction equations are tabulated in this paper. Following choice is possible to introduce as most reliable according to milk indicators: a) for F from M on REF $y = 0.6932x + 1.381$, where $r = 0.752$ and $P \leq 0.001$; b) for P from M on REF $y = 0.9432x + 0.2274$, where $r = 0.949$ and $P \leq 0.001$; c) for L from E on REF $y = 0.8145x + 0.9095$, where $r = 0.87$ and $P \leq 0.001$; d) for log SCC from E on REF $y = 0.8829x + 0.2367$, where $r = 0.94$ and $P \leq 0.001$. The use of equation set in possibilities of MR calculation rear can contribute to favourable ICAR audit of MR in the CR.

SUMMARY

At dairying cost reduction and effort about maintenance of milk recording (MR) range the MR cost decrease is also important. Aim of this paper was to revise the older prediction equations for alternative reduced milk sampling (morning – M or evening – E) for assurance of MR reliable results at twice milking a day (TMD) with balanced interval 12/12. Czech Fleckvieh and Holstein breed model herds were included in the estimation. MR results of last control year in herds 1, 2 and 3 were: 1) 7056 kg of milk per lactation (305 days), 4.05% of fat, 286 kg of fat, 3.65% of protein and 258 kg of protein; 2) 7519, 3.93, 296, 3.45 and 259; 3) 8310 kg, 3.79%, 315 kg, 3.40% and 283 kg. Individual milk samples ($n = 619$ animals) and their values were obtained during TMD (M, E and REF; $n = 1\ 857$ samples) after removal of outliers. Fat (F), protein (P), lactose (L) and somatic cell count (SCC) were analysed (% and 10^3 ml^{-1}). The reference values (REF) of day milking were obtained in regular MR (A4P). M and E milking results were related to the REF by methods of linear and non linear regression. F and P mean values were higher in evening ($4.17 > 3.90$ and $3.74 > 3.63\%$) as compared to morning samples. SKÝPALA and CHLÁDEK (2008) found significant difference for milk proteins between E and M samples (3.27 and 3.24%). Further, they showed significantly higher fat content in evening (4.41%) as compared to morning samples (3.95%). It is in accordance with results of this work. Values of chosen correlation coefficients (at linear relationships) moved from 0.710 ($P \leq 0.001$; F, calculation from E on REF) to 0.949 ($P \leq 0.001$; P, calculation from M on REF). Therefore there were close and very close relationships. There was 50.4% in F case and in P case as many as 90.0% of variability in REF values explainable by variability in values of partial samples which were taken by alternative variants (M and E). Older prediction equations with lower validity because of time and breeding were revised: a) for F from M on REF $y = 0.6932x + 1.381$, where $r = 0.752$ and $P \leq 0.001$; b) for P from M on REF $y = 0.9432x + 0.2274$, where $r = 0.949$ and $P \leq 0.001$; c) for L from E on REF $y = 0.8145x + 0.9095$, where $r = 0.87$ and $P \leq 0.001$; d) for log SCC from E on REF $y = 0.8829x + 0.2367$, where $r = 0.94$ and $P \leq 0.001$. MR cost reduction is possible using method of alternative sampling including corresponding prediction equations.

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