Effect of sires on wide scale of milk indicators in first calving Czech Fleckvieh cows

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Abstract

The possible genetic impact of sire on cattle populations, herd milk yield and milk traits (fat and protein) have been described in the literature along with its impact on some milk indicators (MIs) as somatic cell count, urea and ketones. There is a dearth of information on the impact on a series of other MIs (physical, chemical, health, technological). The goal of this study was to assess the possible effect of sire on a wide range of MIs including technological properties in Czech Fleckvieh to suggest future possible breeding trends. A series of MIs (n=37) was investigated in individual milk samples (MSs). 191 effective daughters (MSs) were included. The sire groups (n=13) were well balanced in terms of herd, lactation stage and sampling season. Only sires with >5 daughters were ranked. A linear model of analysis variance with the fixed effects, sire and combined factor (herd \times year \times season) was used. 19 MIs as log count of streptococci in fermentation ability of milk (log FAM–CS), FAM–CS, log total fine microflora count in FAM (log FAM–TCM), FAM–TCM, solids non fat (SNF), iodine content, citric acid (CA), titration acidity in FAM, lactose (L), crude protein (CP), true protein (TP), casein (CAS), dry matter, Mg and P content, milk alcohol stability, electrical conductivity (EC), titration acidity, casein numbers (for CP and TP), log count of lactobacilli in FAM (log FAM–CL), FAM–CL and pH in FAM were influenced by sire (P<0.05). However SNF, CA, L, CAS and perhaps EC could be newly reflected as information for genetic improvement of dairy cattle with connection to dairy milk recovery and cow health. CA (10.08 \pm 1.92 mmol× l^{-1}) deserves special attention. The model variability explanation moved from 6.97 (SCC) over 29.51 (CA) to 48.32 % (pH) for MIs. This is one of few studies to assess the impact of sire over a wide range of MIs and the results warrant careful evaluation and further study.

Keywords: sire, Fleckvieh, first calving cow, milk, composition indicators, casein, health indicators, citric acid, physical and technological properties, electrical conductivity, milk fermentation ability

Zusammenfassung

Einfluss der Bullen auf eine breite Skala von Milchindikatoren bei erstkalbenden Töchtern des Tschechischen Fleckviehs

Der mögliche genetische Bulleneinfluss auf die Milchleistung und Milchinhaltstoffe (Fett und Eiweiß) sowie Milchindikatoren wie somatische Zellzahl, Harnstoff oder Ketone wurden hinlänglich in der Literatur beschrieben. Über weitere chemische, physikalische, gesundheitliche oder technologische Milchindikatoren liegen weniger Informationen vor. Ziel vorliegender Untersuchung war die Prüfung des Vätereinflusses auf die letztgenannten Milchindikatoren beim Tschechischen Fleckvieh für die Gestaltung künftiger Zuchtziele. An individuellen Milchproben von 191 effektiven Töchtern wurden 37 Milchindikatoren erfasst. Die 13 einbezogenen Gruppen von Kühen waren hinsichtlich der Einflüsse Herde, Laktationsstadium und Jahreszeit der Probenname ausgeglichen. Bewertet wurden nur Bullen mit mehr als fünf Töchtern. Für die Varianzanalyse fand ein lineares Modell Anwendung welches neben den fixen Effekten Bulle die kombinierten Faktoren Herde, Jahr und Jahreszeit einschloss. Für folgende Milchindikatoren wurde ein signifikanter (P<0,05) Bulleneinfluss nachgewiesen: Streptokokkenzahl (log FAM-CS), FAM-SC, Mikroflorazahl in der FAM (log FAM-TCM), FAM-TCM, fettfreie Trockensubstanz (SNF), Jodgehalt, Zitronensäuregehalt (CA), Titrationssäuregehalt in FAM, Laktosegehalt (L), Roheiweißgehalt, (CP), Reineiweißgehalt (TP), Kasein (CAS), Trockenmasse, Mg- und P-Gehalt, Milchalkoholstabilität, elektrische Leitfähigkeit EC), Tritationssäuregehalt (Milchsäurezahl), Kaseinzahlen für CP und TP, Laktobazilluszahl in der FAM (log FAM-CL), FAM-CL und pH in der Fam. Als neue Informationsquellen würden sich für eine züchterische Verbesserung der Milchkuhpopulation nur SNF, CA, L, CAS und evt. EC im Zusammenhang mit Milchqualität und Kuhgesundheit eignen. Der CA Wert (10,08 \pm 1,92 mmol \times I⁻¹) muss gesondert betrachtet werden. Die Modellvariabilität der Milchindikatoren bewegte sich um Werte von 6.97 (SCC) über 29,51 (CA) bis 48,32 (pH). Die vorliegende Studie ist eine der wenigen welche den Bulleneinfluss auf eine Vielzahl von Milchindikatoren untersucht deshalb sind die Ergebnisse mit Bedacht zu bewerten und weitere Untersuchungen erforderlich.

Schlüsselwörter: Bulle, Fleckvieh, erstkalbende Kuh, Milch, Milchzusammensetzung, Kasein, Gesundheitsindikatoren, Zitronensäure, Milcheigenschaften physikalisch technologisch, elektrische Leitfähigkeit, Milchfermentationsfähigkeit

Introduction

Milk yield and milk composition (milk indicators, Mls), reproduction performance (Bezdíček *et al.* 2007, Parland *et al.* 2007), health status (Hinrichs *et al.* 2006) and other functional traits are important data for the genetic improvement of dairy cattle (Bergfeld & Klunker 2002, Distl 2001). The genetic impact of sire on cattle populations, herd milk yields and some Mls have been desribed in a number of papers (Thompson *et al.* 2007a, 2007b, Biedermann *et al.* 2003, 2004, Bezdíček *et al.* 2008). These are usually called milk traits and most of the research has been carried out for such milk traits as milk fat and protein yield and percentage, dry matter (Schutz *et al.* 1990, Yazgan *et al.* 2010) and also somatic cell count (Schutz *et al.* 1990, Xu *et al.* 2006) as indicators of mammary gland health status. Some authors have also studied the effects of genetics on health and nutrition as Mls such as lactose, urea and ketones (Gravert *et al.* 1991, Wood *et al.* 2004, Miglior *et al.* 2006, Stoop *et al.* 2007). From a genetic point of view, there is still a dearth of information on a range of other important Mls. The goal of this

study was therefore to assess the possible effects of sire on a wide range of important milk indicators including technological properties in Czech Fleckvieh to suggest a future possible trend in breeding work.

Materials and methods

Animals and milk samples

Individual milk sample (MSs) collection was carried out at five commercial dairy farms of the Czech Fleckvieh (CF) breed. Only the first calved dairy cows were sampled from 90 to 180 days in milk. The sampling period was two years and 191 MSs were collected. The dairy cows were kept in binding free stables with milking parlours and all were milked twice a day. The dairy cow nutrition was composed of TMR, which is typical for the country conditions and consisted of: maize silage; alfalfa silage; trifolium silage; whole spindle maize silage (LKS); brewery draff; alfalfa hay; concentrates. TMR and concentrates were fed according to milk yield and standard demands.

Analyses of the individual milk samples

The list of analyses is shown in Table 1 for 37 MIs. The analyses were carried out according to the following milk analytical procedures. The fat (F), L and SNF indicators were measured using MilkoScan 133B (Foss Electric, Denmark) calibrated according to reference method results (standard CSN 57 0536 and CSN 57 0530). The protein fractions CP, TP and CAS (N×6.38) were determined by Kjeldahl's method using the instrument line Tecator with Kjeltec Auto Distillation unit 2 200 (Foss-Tecator AB, Sweden) according to CSN 57 0530. The macro- and microelement milk contents were investigated by atom absorption spectrophotometry using a Spectrometer SOLAAR S4 and 6F S97 Thermo Elemental (England). The SCC was determined using Fossomatic 90 instrument (Foss Electric, Denmark) according to CSN EN ISO 13366–2. The U was determined by spectrophotometry at 420 nm wavelength. The specific reaction solution was prepared as a sour mixture with the p-dimethylaminobenzaldehyde (Hanuš et al. 1995) using Spekol 11 instrument (Carl Zeiss Jena, Germany). The AC was investigated by spectrophotometry at 485 nm wavelength. The AC was absorbed in alkali solution of KCl with salicylaldehyde after to 24 hours microdiffusion (O'Moore 1949, Voitíšek 1986) using Spekol 11. The CA was determined by spectrophotometry at 428 nm wavelength. Milk was coagulated by trichloracetic acid and the adventitious filtrate then allowed to react with pyridine and acetanhydride (30 min at 32 °C). Used instrument was Spekol 11 (Hanuš et al. 2009). The MFP values were analysed using a top cryoscope Cryo-Star automatic Funke-Gerber (Germany) which was calibrated by standard NaCl solutions (Funke-Gerber). The EC was measured using OK 102/1 (Radelkis, Hungary) conductometer at 20 °C (in mS×cm⁻¹) with bell glass electrode which was calibrated by salt (KCI) solution (10.2 mS×cm⁻¹). The active acidity (pH) was measured using the pH-meter CyberScan 510 (Eutech Instruments) which was calibrated by buffer solutions (pH 4.0 and 7.0) at 20 °C. The TA was measured by milk titration (100 ml) using alcaline solution according to CSN 57 0530. The AS was determined with the help of the milk titration (5 ml) by 96% ethanol (ml) to the creation of protein precipited flakes. The FAM–CL, –CS and –TCM (carried out according to standard ON 57 0534 by slightly modified

Table 1

procedure with thermophilic yoghurt culture YC–180–40–FLEX=*Streptococcus thermophilus, Lactobacillus delbrueckii subsp. lactis* and *L. d. subsp. bulgaricus*) were investigated by calculating of the colony forming units (CFU) using the classical plate cultivation method (at 30 °C for 72 h) with GTK M (Milcom Tabor) agar according to CSN ISO 6610.

Abbreviation	Milk indicator	Unit	Note
F	fat	g×100g ⁻¹ ;%	
L	lactose	q×100q ⁻¹ ;%	monohydrate
СР	crude protein	q×100q ⁻¹ ;%	
ТР	true protein	q×100q ⁻¹ ;%	
CAS	casein	g×100g ⁻¹ ;%	
WP	whey protein	q×100q⁻¹;%	
NPN	non protein nitrogen matter	g×100g ⁻¹ ;%	
URN	urea nitrogen ratio in NPN	%	
SNF	solids non fat	g×100g ⁻¹ ;%	
DM	dry matter	g×100g ⁻¹ ;%	total solids
Ca, P, Na, Mg, K	-	mg×kg⁻¹	macroelements
1		µg×l ^{−1}	microelement
SCC	somatic cell count	thousand×ml ⁻¹	
U	urea	mg×100ml⁻¹	
AC	acetone	mg×l ⁻¹	
CA	citric acid	mmol×l ⁻¹	
F/CP	fat/crude protein ratio		
AS	alcohol stability	ml	thermostability replacement
TA	titration acidity	ml	Soxhlet–Henkel
EC	conductivity	mS×cm ⁻¹	electrical conductivity
рН	actual acidity		
MFP	milk freezing point	°C	
CAS-CP	casein number	%	on CP basis
CAS-TP	casein number	%	on TP basis
RCT	cheeseability	second	rennet coagulation time
CQ	cheeseability	class	curd cake quality
CF	cheeseability	cm	cheese curd firmness
WV	cheeseability	ml	whey volume
FAM-T	fermentation ability	ml	milk yoghurt test
FAM-pH	yog. actual acidity		milk yoghurt test
FAM-CL	<i>lactobacilli</i> count	CFU×ml ⁻¹	yoghurt, colony forming unit
FAM-CS	streptococci count	CFU×ml ⁻¹	yoghurt, colony forming unit
FAM-TCM	all microorganisms	CFU×ml ⁻¹	fine fermenting microorg.
FAM-RSL	ratio		yog. streptococci/lactobacilli

List of abbreviations of milk indicators (MIs) with their units

TA: ml 0.25 mol×l⁻¹ NaOH solution for the titration of 100 ml of milk (CSN 57 0530), AS: consumption of 96% ethanol in ml to protein coagulation in 5 ml of milk, CQ: subjective estimation determined by inspection and touch from 1st (excellent) to 4th (poor) class, CF: depth of penetration of the corpuscle falling into curd cake in the standard way expresses the opposite relationship to firmness, WV: whey which was ejected during rennet curd cake creation for 60 min, FAM–T: with microbial culture (by titration acidity in ml of 0.25 mol×l⁻¹ NaOH×100ml⁻¹); all the previous parameters at FAM were measured after the yoghurt test fermentation.

Design of the investigation and statistical procedures

13 sires with more than five daughters were included in the data set for statistical evaluation. It means, 191 effective daughters (MSs) were included into investigation. The sire daughter result groups were well balanced in terms of herd, lactation stage and sampling season. Analysis of variance with fixed effects as sire and lactation stage was used for statistical evaluation of data set according to model as follows:

$$y_{ij} = \mu + hys_i + s_j + e_{ij}$$
 (1)

where *y* is the investigated milk indicator, μ is the general average, *hys* is the herd × year × season effect (combined effect including the impact of herd, year of calving and season of calving) for *i* from 1 to 6 combinations (this effect used for elimination of major part of systematic environmental variation), *s* is the sire effect for *j* from 1 to 13 (Figure 1) sires and *e* is the random effect.

The SAS v9 programme package was used for the calculation. Means and GLM procedures were performed.

As the usual evaluation of milk yield traits was not main object of this evaluation fat and protein yields were not calculated and only fat and protein percentage were evaluated according to analytical results. The other milk indicators were the main goal of investigation. Therefore, SCC, AC and hygienic indicator values (FAM–CL, FAM–CS, FAM–TCM; Table 1) were logarithmically transformed (log) on a decimal basis because of lack of normal frequency distribution in most cases (Shook 1982, Reneau 1986, Meloun & Militký 1994) and after that geometrical averages were also used in results evaluation. This data set had a smaller range than is usual for genetic evaluation of milk traits in the population. In contrast this result set was quite large in terms of evaluating such a wide spectrum of MIs which is not often case.

Results and discussion

Main statistic characteristics of milk indicators

The statistics for the MIs are shown in Table 2. The means and variability are mostly very comparable to our previous results in CF (Hanuš *et al.* 2007) and not very different from results in Holstein (H) (Janů *et al.* 2007). Differences (compared to CF) are probably connected to first lactation effectshere for instance for L and SNF contents which are markedly higher (5.11>4.96 % and 9.11>8.91 %; than CF results; Hanuš *et al.* 2007). Some differences (compared to H) are probably connected, apart from to first lactation also with breed and milk yield (H higher) effects for example especially for main protein fractions. The CP, TP and CAS contents are markedly higher for CF: 3.42>3.24 %; 3.23>3.07 %; 2.75>2.57 % (compared to H results; Janů *et al.* 2007).

		Maan	Chan da val da viatia		Massimo	
variable	n	Mean	Standard deviatio	n Minimum	Maximum	
F	191	3.91	0.548	2.28	5.67	
L	191	5.11	0.225	3.32	5.48	
CP	191	3.42	0.252	2.82	4.20	
ТР	191	3.23	0.248	2.69	4.00	
CAS	191	2.75	0.247	1.81	3.40	
WP	191	0.48	0.099	0.28	1.11	
NPN	191	0.191	0.053	0.09	0.52	
URN	191	58.68	15.87	16.03	98.47	
SNF	191	9.11	0.325	7.70	9.83	
DM	191	13.02	0.681	10.62	14.82	
Ca	144	1 340.6	199.1	803.0	1 782.0	
Р	142	1 178.3	166.1	824.0	1 659.0	
Na	144	383.8	101.6	230.0	1 100.0	
Mg	144	117.8	11.6	88.3	145.3	
К	144	1676.0	138.6	1 183.0	1 989.0	
	120	333.1	124.8	116.0	690.0	
SCC	191	184	623	10	8011	
log SCC	191	1.90794	0.44366	1.0	3.90370	
U	191	36.63	6.40	20.87	53.86	
AC	191	3.47	3.03	0.01	31.69	
log AC	191	0.35306	0.55154	-2.0	1.50090	
CĂ	144	10.08	1.92	4.55	14.91	
F/CP	191	1.148	0.171	0.62	1.69	
AS	191	0.435	0.174	0.17	1.02	
TA	191	8.23	1.03	4.93	11.22	
EC	191	3.88	0.388	3.11	5.88	
рН	191	6.67	0.146	5.53	7.12	
MFP	191	-0.52873	0.00731	-0.54830	-0.50980	
CAS-CP	191	80.41	3.09	59.15	86.54	
CAS-TP	191	85.18	3.111	61.99	90.48	
RCT	191	132.0	61.6	30.0	510.0	
CQ	191	2.31	1.029	1	4	
CF	191	1.77	0.167	0.5	2.0	
WV	191	33.32	3.179	13.0	40.0	
FAM-T	191	31.85	3.299	22.43	42.54	
FAM-pH	191	4.89	0.182	4.46	5.30	
FAM-CL	191	24 623 560	13 539 397	3600000	84000000	
log FAM–CL	191	7.32879	0.24036	6.55630	7.92430	
FAM-CS	191	643 769 634	206 765 642	200 000 000	1600 000 000	
log FAM–CS	191	8.78589	0.14517	8.30100	9.20410	
FAM-TCM	191	668 393 194	212 821 965	214000000	1642000000	
log FAM–TCM	191	8.80265	0.14344	8.33040	9.21540	
FAM-RSL	191	33.05	20.61	8.82	183.33	

Table 2
Main statistical characteristics of milk indicators in Czech Fleckvieh first calving dairy cows

Mean: arithmetical mean

Sire effects on milk indicators in general

Statistic model efficiency and the significance of sire and combined effects for MIs are shown in Table 3. The model variablity explanation moved from 6.97 (SCC) over 29.51 (CA) to 48.32 % (pH) along individual MIs. According to evaluation of model determination coefficients and model significance and significance of both effects (sire and combined effect) it is possible to mark MIs which are important for interpretation of incidental sire impacts (Table 3, bold letters). The most important were L, CP, TP CAS, SNF, DM, P, Mg, I, CA, AS, TA, EC, CAS–CP, FAM–T, log FAM–CS and log FAM–TCM in this data set. There was a lower explanation of variability by model, no significant effect of sire and/ or too essential impact of combined effect (environmental conditions) for MIs. The model explained most of the variability for pH, CF, log FAM–CS, log FAM–TCM, AC, SNF, I, CA and FAM-T but only at log FAM–CS, log FAM– TCM, SNF, I, CA and FAM-T was this possible to explain by sire effect.

Sire effects on main milk composition and nitrogen fraction indicators

Fat content, which is normally included in genetic improvement programmes was not influenced by sire in this smaller data set, probably due to the greater variability of this MI. CP, which is also normally included in genetic improvement, TP and CAS was significantly influenced by sire (Table 3). The most significant effect was at CAS (Figure 1, CAS), which could be newly routinely determined in dairy analytical systems (Broutin 2006 a; MIR-FT under certain circumstances) and in preference included in dairy herd improvement programmes. Lactose content as an indicator of udder health (Hanuš *et al.* 1993), which is connected with high milk yield (higher L; Janů *et al.* 2007, Hanuš *et al.* 2007) was influenced significantly by sire (Table 3). Miglior *et al.* (2006) reported marked difference between Ayrshire and H dairy cows (L 4.49 and 4.58 %; anhydride) in Canada, probably as a genetic factor. SNF and DM were also influenced by sire (Table 3). It was logically in links with protein fractions and L.

Tab	le 3
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SAS GLM procedure results for milk indicators in terms of sire and combined (Com., herd×year×season) effect in Czech Fleckvieh first calving dairy cows

Variable	Model R ²	Model F	Model sign.	Sire F	Sire sign.	Com. F	Com. sign.
F	10.55	1.20	0.2684	1.31	0.2170	0.94	0.4565
L	22.51	2.96	0.0002	2.85	0.0013	3.20	0.0086
СР	19.76	2.51	0.0014	3.14	0.0005	0.98	0.4306
ТР	20.57	2.64	0.0008	3.48	0.0001	0.61	0.6927
CAS	21.24	2.74	0.0005	3.84	<0.0001	0.12	0.9866
WP	11.97	1.38	0.1492	1.28	0.2365	1.64	0.1508
NPN	10.22	1.16	0.3037	1.13	0.3366	1.22	0.3018
URN	13.85	1.64	0.0598	1.75	0.0605	1.37	0.2394
SNF	30.90	4.55	<0.0001	6.16	<0.0001	0.70	0.6259
DM	17.44	2.15	0.0071	2.75	0.0019	0.71	0.6179
Ca	10.33	0.86	0.6209	0.99	0.4667	0.56	0.7292
Р	18.54	1.67	0.0563	2.03	0.0268	0.82	0.5382
Na	18.65	1.71	0.0485	1.73	0.0671	1.66	0.1485
Mg	25.32	2.53	0.0017	3.10	0.0007	1.17	0.3265
ĸ	17.12	1.54	0.0899	1.80	0.0546	0.93	0.4667
I	30.85	2.70	0.0010	3.63	0.0002	0.49	0.7824
SCC	6.97	0.76	0.7335	0.45	0.9428	1.53	0.1842
log SCC	19.31	2.44	0.0020	1.58	0.1026	4.50	0.0007
U	13.65	1.61	0.0665	1.78	0.0540	1.19	0.3179
AC	31.57	4.69	<0.0001	1.60	0.0942	12.11	<0.0001
log AC	13.54	1.59	0.0701	1.08	0.3780	2.82	0.0177
CA	29.51	3.13	0.0001	3.79	<0.0001	1.53	0.1848
F/CP	12.82	1.50	0.1007	1.61	0.0918	1.22	0.3034
AS	21.29	2.75	0.0005	3.66	<0.0001	0.59	0.7092
ТА	20.22	2.58	0.0010	2.87	0.0012	1.88	0.0994
EC	27.80	3.92	<0.0001	5.15	<0.0001	0.96	0.4452
рН	48.32	9.52	<0.0001	1.63	0.0876	28.45	<0.0001
MFP	12.58	1.46	0.1131	1.45	0.1457	1.49	0.1969
CAS-CP	16.59	2.02	0.0122	2.03	0.0244	2.02	0.0788
CAS-TP	14.82	1.77	0.0351	2.01	0.0261	1.21	0.3088
RCT	13.17	1.91	0.0283	1.64	0.0836	3.49	0.0325
CQ	28.42	4.04	<0.0001	1.28	0.2365	10.68	<0.0001
CF	34.00	5.24	<0.0001	0.60	0.8385	16.38	<0.0001
WV	24.47	3.30	<0.0001	1.40	0.1681	7.84	<0.0001
FAM-T	29.41	4.24	<0.0001	4.69	<0.0001	3.15	0.0096
FAM-pH	18.00	2.23	0.0049	1.84	0.0452	3.18	0.0090
FAM-CL	17.04	2.09	0.0092	2.61	0.0032	0.84	0.5226
log FAM–CL	17.69	2.19	0.0060	2.49	0.0049	1.45	0.2093
FAM-CS	25.35	3.46	<0.0001	4.21	<0.0001	1.63	0.1536
log FAM–CS	31.72	4.73	<0.0001	5.94	<0.0001	1.82	0.1120
FAM-TCM	25.94	3.56	<0.0001	4.36	<0.0001	1.66	0.1459
log FAM–TCM	31.67	4.72	<0.0001	5.93	<0.0001	1.80	0.1155
FAM-RSL	22.40	2.94	0.0002	1.65	0.0818	6.03	<0.0001

 R^2 : coefficient of determination, %, sign.: statistical significance, F: fat value, **bold letters** in column variable mean important for interpretation in terms of sire effect, **bold letters** in columns R^2 (>25%) and sign. mean the most important for interpretation; *script letters* in columns R^2 (<15%) and sign. mean no important for interpretation.



Sires: 1, HEL 8; 2, HG 109; 3, HG 141; 4, MKM 221; 5, MKM 229; 6, MOR 40; 7, MOR 45; 8, MOR 51; 9, MOR 59; 10, RAD 121; 11, RAD 64; 12, REZ 327; 13, UF 59. Columns: black *P*<0.05; dash *P*<0.10; empty *P*>0.10.

Figure 1

Graphical rendering of sire effects on selected milk indicators in Czech Fleckvieh first calving dairy cows

Sire effects on some milk mineral components

Microelement I and macroelements as Mg and P were significantly influenced by sire (Table 3) in this data set. Total P is logically partly connected with casein fraction. Owing to high variability there could be a systematic interference effect caused by the presence (diffusion effect) and or absence of iodine from udder teat disinfection in daughters for I results (Table 2) in practice. Mg and I are minerals important to human nutrition from milk products in connection with nerve and overall metabolism. However in practical terms including these minerals into dairy cattle improvement is improbable.

Sire effect on dairy cow health state milk indicators

SCC is a well-known indicator of mammary gland health in lactating mammals (Shook 1982, Reneau 1986, Amin 2001, Amin *et al.* 2002, Kühn *et al.* 2008). The geometric average of SCC was 81 thousand×ml⁻¹ (Table 2) and this confirmed that only relatively healthy dairy cows (clinical mastitis free) were used in this evaluation. In this paper the SCC was not influenced by sire effect (Table 3) although SCC is evaluated routinely in terms of mastitis resistance determination in dairy cattle populations, for instance as sire herd book values. Nevertheless, the coefficient heritability for somatic cell score was also low 0.1 (Schutz *et al.* 1990). The log SCC was influenced by combined environmental effect in this paper which is practically quite logical (Table 3).

Milk urea content is good indicator of nitrogen matter/energy metabolism in dairy cows (Larson et al. 1997, Panicke et al. 2000, Godden et al. 2001 a, 2001b, Mottram & Masson 2001, Rajala-Schultz et al. 2001, Guo et al. 2004, Jílek et al. 2006, Miglior et al. 2006, Zhai et al. 2006, Stoop et al. 2007, Bezdíček et al. 2009, Řehák et al. 2009) mostly with negative relation to reproductive performance and longevity. Urea was not significantly influenced by either used model factors in this paper (Table 3; $36.6\pm6.4 \text{ mg}\times100 \text{ ml}^{-1}$). Also the model variability explanation was lower. Nevertheless, the sire effect was almost significant at alpha 0.05 (P=0.054; Table 3). The possibility of including U into routine milk recording and dairy cattle improvement in terms of nitrogen matter utilization from feeding rations could be a topic of further research. Miglior et al. (2006) mentioned lower milk U nitrogen in H dairy cows than Ayrshire (11.11<12.20 mg×100ml⁻¹). Lower U was found along with lower milk yield between breeds. This is in good accordance with previous results (Hanuš et al. 2007 and Janů et al. 2007), where U was lower in H breed than CF and also was lower along with lower milk yield within both breeds and between breeds. Also Godden et al. (2001 b) found a positive nonlinear association between milk urea and milk yield and energy corrected milk in Ontario H cattle. Stoop et al. (2007) reported milk U nitrogen heritability 0.14, which is lower. Nevertheless, they allowed for the possibility of influencing urea by through genetic selection apart from herd practice.

Milk acetone was evaluated as a suitable indicator of animal energy metabolism. The higher AC the lower energy support or ketosis according to a number of authors (Gravert *et al.* 1986, 1991, Enjalbert *et al.* 2001, Mottram *et al.* 2002, Wood *et al.* 2004). AC was not influenced by sire in this data set although significance was quite close to set limits of probability (P=0.0942; Table 3; geometric mean 2.25 mg×l⁻¹). In contrast, a significant impact was found for combined environmental factor (Table 3). Nevertheless, Gravert *et al.* (1991) in cows with an increased AC in the first third of lactation (0.25 mmol.l⁻¹) a negative correlation (r=–0.47 and –0.42, resp.) to the energy quantity received through fodder and also to milk yield (r=–0.30). They reported a heritability coefficient for milk AC 0.30 which was similar to the milk yield coefficient. Therefore, milk AC content was recommended as assessment of energy balance and to be included in milk recording and breeding value determination in terms of genetic control of energy nutrient utilization. Wood *et al.* (2004) reported heritability of AC less than 1%, therefore they mentioned a genetic evaluation based on milk acetone recording on a monthly basis as having little use as a selection tool to decrease the incidence of ketosis.

Milk citric acid could be also a good indicator of animal energy metabolism (low CA means low energy maintenance) according to more papers (Khaled *et al.* 1999, Baticz *et*

al. 2002, Garnsworthy *et al.* 2006, Kubešová *et al.* 2009). The lower milk CA the higher the energy deficiency and vice versa. The physiological range is between 8 and 10 mmol×l⁻¹. Garnsworthy *et al.* (2006) confirmed the hypothesis that variation in CA with stage of lactation was related to de novo synthesis of fatty acids and that the relationship was independent of diet and milk yield. This is promising for genetical possibilities. CA (10.08±1.92 mmol×l⁻¹) was significantly influenced only by sire along good model variability explanation (Table 3). This is the most interesting result in this work (Figure 1, CA). The CA as an energy metabolism MI could be suitable for milk recording and dairy herd improvement as it is practical. This deserves special attention from the genetic evaluation point of view. Similarly metioned by Gravert *et al.* (1991) for milk acetone and control of feeding rations (energy nutrients) in dairy cows. Also routine series CA analyses are now accessible for effective milk laboratory work (Hanuš *et al.* 2009).

The F/CP ratio could be a suitable indicator of animal energy metabolism (Geishauser & Ziebell 1995, Heuer *et al.* 1999, 2001). The higher the F/CP, the lower the metabolism energy support and possible ketosis. In contrast, a low F/CP ratio could provide information on low cow maintenance by structure fiber. F/CP ratio was not influenced by either sire or combined effect in this material.

Sire effects on physical indicators

Out of the physical MIs investigated the EC as a possible indicator of udder health (Hanuš *et al.* 1993) was statistic significantly influenced only by sire (Table 3, Figure 1, EC) and the model variability explanation was quite successful in this case. EC included into routine milk recording and dairy herd improvement is feasible. There is also a possibility of routine series measurements in dairy cow herds. The pH and MFP were not influenced by sire. However, pH was significantly influenced by combined environmental effect.

Sire effects on milk technological properties

Milk indicators as AS, TA, CAS–CP (Figure 1), CAS–TP, FAM–T, log FAM–CS (Figure 1), FAM– CS, log FAM–TCM (Figure 1), FAM–TCM, log FAM–CL, FAM–CL and FAM–pH (Table 3) were influenced by sire significantly. This refers to almost all MIs of milk fermentation ability. The cheeseability indicators as RTC, CQ, CF and WV, which were significantly influenced by milk yield and differed between cattle breed (CF and H; Hanuš *et al.* 2007, Janů *et al.* 2007) were not influenced by sire in this data set (Table 3). On the other hand these were significantly influenced by combined effect.

This paper is one of few studies to assess sire impact over such wide range of MIs. Therefore the results for the technological properties need to be evaluated carefully. For example apropos fermentation ability, there are a number of interference interactions among milk components, properties and fine culture activities. Nevertheless, out including these technological properties in dairy cattle improvement is improbable in practice with the exception of AS and TA. However, their routine series determination is problematic. In conclusion 19 variables (Table 3) as log FAM–CS, FAM–CS, log FAM–TCM, FAM–TCM, SNF, I, CA, FAM-T, L, CP, TP, CAS, DM, Mg, P, AS, EC, TA, CAS–CP, CAS–TP, log FAM–CL, FAM–CL and FAM–pH from 37 MIs (Table 2) were significantly influenced by sire effect in this data set.

Of these, only the SNF, CA, L, CAS and perhaps EC reflect new information for the genetic improvement of dairy cattle with connection to dairy milk recovery and cow health. Care is necessary in the results interpretation. Further studies are also necessary. Nevertheless, the importance of new genetic assessments for a whole series of MIs in terms of future cattle breeding work is currently increasing. This refers to rapid dissemination of modern effective technologies and milk analytical methods like NIR-FT and MIR-FT (near and mid infra-red spectrophotometry with Fourier's transformations) and other methods mostly on the basis of biosensors which are able to measure new MIs like casein, citric acid, urea, acetone (ketones), free fatty acids, milk freezing point and electrical conductivity with high efficiency (Koops *et al.* 1989, Hansen 1999, Heuer *et al.* 2000, Tsenkova *et al.* 2000, Kukačková *et al.* 2000, Broutin 2006 a, b, Mottram & Masson 2001, Mottram *et al.* 2002, Jankovská & Šustová 2003, Miglior *et al.* 2006, Roos *et al.* 2006, Bijgaart 2006, Šustová *et al.* 2007, Hanuš *et al.* 2008). These results could contribute to reliable data about milk indicators for official milk recording for possible inclusion into breeding and dairy cattle improvement. Possible trends were indicated in this paper. It could be used for next research trend in dairy cattle genetic improvement procedure.

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References

- Amin AA (2001) Lactation and sample test-day multi-trait animal model for genetic evaluation of somatic cell scores in Hungarian Holstein Friesian crossbreeds. Arch Tierz 44, 263-275
- Amin AA, Gere T, Kishk WH (2002) Genetic and environmental relationship among udder conformation traits and mastitis incidence in Holstein Friesian into two different environments. Arch Tierz 45, 129-138
- Baticz O, Tömösközi S, Vida L (2002) Concentrations of citrate and ketone bodies in cow's raw milk. Periodica Polytechnica Ser Chem Eng 46, 93-104
- Bergfeld U, Klunker M (2002) Importance of functional traits in cattle breeding and possibilities to improve by breeding. Arch Tierz 45, Special Issue, 60-67 [in German]
- Bezdíček J, Hanuš O, Bjelka M, Dufek A, Jedelská R, Kopecký J (2009) Analyse of the relationship between milk components and reproduction in the Czech Fleckvieh first-calf cows. Acta univ agric et silvic Mendel Brun LVII, 1, 13-26
- Bezdíček J, Šubrt J, Filipčík R (2008) Inbreeding depression of milk traits in Holstein cows in the Czech Republic. Arch Tierz 50, 415-425
- Bezdíček J, Šubrt J, Filipčík R, Bjelka M, Dufek A (2007) The effects of inbreeding on service period and pregnancy length in Holsteins and Czech Fleckviehs after the first calving. Arch Tierz 50, 455-463
- Biedermann G, Waldmann S, Maus F (2003) Genetic analysis of the population of Hinterwald cattle. Arch Tierz 46, 307-319 [in German]
- Biedermann G, Ott B, Rübesam K, Maus F (2004) Genetic analysis of the population of Vorderwald cattle. Arch. Tierz. 47, 141-153 [in German]
- van den Bijgaart HJCM (2006) New applications of mid-infra-red spectrometry for the analysis of milk and milk products. 2 Free fatty acids. IDF Bulletin, 406, 22-28
- Broutin PJ (2006 a) New applications of mid-infra-red spectrometry for the analysis of milk and milk products. 1 Casein. IDF Bulletin, 406, 2-21

- Broutin PJ (2006 b) Use of highly accurate enzymatic method to evaluate the relationship between milk urea nitrogen and milk composition and yield on bulk and individual milk samples. 35th ICAR Session, Kuopio, Finland
- CSN 57 0530: Methods for testing of milk and milk products. Standard CNI Prague, 1972 [in Czech]
- CSN 57 0536: Determination of milk composition by mid-infrared analyser. Standard CNI Prague, 1999 [in Czech]
- CSN EN ISO 13366-2 (57 0531): Milk Enumeration of somatic cells Part 2: Manual for control of fluoro-optoelectronic instrument. Standard CNI Prague, 2007 [in Czech]
- CSN ISO 6610: Milk and milk products. Enumeration of colony-forming units of micro-organisms. Colonycount technique at 30 °C. Standard CNI Prague, 1996 [in Czech]
- Distl O (2001) Implication of health traits in breeding of dairy cattle. Arch Tierz 44, 365-380 [in German]
- Enjalbert F, Nicot MC, Bayourthe C, Moncoulon R (2001) Ketone bodies in milk and blood of dairy cows: Relationship between concentrations and utilization for detection of subclinical ketosis. J Dairy Sci 84, 583-589
- Garnsworthy PC, Masson LL, Lock AL, Mottram TT (2006) Variation of milk citrate with stage of lactation and de novo fatty acid synthesis in dairy cows. J Dairy Sci 89, 1604-1612
- Geishauser T, Ziebell KL (1995) The fat/protein ratio in the milk of cattle herds with the occurence of abomasal displacement. Dtsch Tierärztl Wschr 102, 469-494 [in German]
- Godden SM, Kelton DF, Lissemore KD, Walton JS, Leslie KE, Lumsden JH (2001 a) Milk urea testing as a tool to monitor reproductive performance in Ontario dairy herds. J Dairy Sci 84, 1397-1406
- Godden SM, Lissemore KD, Kelton DF, Leslie KE, Walton JS, Lumsden JH (2001 b) Factors associated with milk urea concentrations in Ontario dairy cows. J Dairy Sci 84, 107-114
- Gravert HO, Jensen E, Hefazian H, Pabst K, Schulte-Coerne H (1991) Environmental and genetic effects on the acetone content in milk. Züchtungskunde 63, 42-50 [in German]
- Gravert HO, Langer R, Diekmann L, Pabst K, Schulte-Coerne H (1986) Ketone bodies in milk as indicators for energy balance in dairy cows. Züchtungskunde 58, 309-318 [in German]
- Guo K, Russek-Cohen E, Varner MA, Kohn RA (2004) Effects of milk urea nitrogen and other factors on probability of conception of dairy cows. J Dairy Sci 87, 1878-1885
- Hansen PW (1999) Screening of dairy cows for ketosis by use of infrared spectroscopy and multivariate calibration. J Dairy Sci 82, 2005-2010
- Hanuš O, Frelich J, Janů L, Macek A, Zajíčková I, Genčurová V, Jedelská R (2007) Impact of different milk yields of cows on milk quality in Bohemian Spotted cattle. Acta Vet Brno 76, 563-571
- Hanuš O, Gabriel B, Genčurová V, Žváčková I (1993) Lactose content in cow milk in the first third of lactation according to some indicators of secretion disorder of mammary gland. Živoč Výr/Czech J Anim Sci 38, 131-138 [in Czech]
- Hanuš O, Hulová I, Genčurová V, Štolc L, Kučera J, Kopecký J, Jedelská R, Motyčka Z (2009) Result interpretation of experimental calibration for milk citric acid determination via infrared spectroscopy (MIR-FT). Acta univ. agric. et silvic. Mendel Brun, ISSN 1211-8516, LVII, 77-101 [in Czech]
- Hanuš O, Jílek M, Ficnar J, Beranová A, Jedelská R, Havlíčková K, Míčová Z (1995) Ways of preparing standards for calibration of indirect methods of determination of urea concentration in milk. Živoč Výr/Czech J Anim Sci 40, 441-451 [in Czech]
- Hanuš O, Vegricht J, Frelich J, Macek A, Bjelka M, Louda F, Janů L (2008) Analyse of raw cow milk quality according to free fatty acids contents in the Czech Republic. Czech J Anim Sci 53, 17-30
- Heuer C, Luinge HJ, Lutz ETG, Schukken YH, van der Maas JH, Wilmink H, Noordhuizen JPTM (2000) Determination of acetone in cow milk by Fourier transform infrared spectroscopy for the detection of subclinical ketosis. J Dairy Sci 83, 575-582
- Heuer C, Schukken YH, Dobbelaar P (1999) Postpartum body condition score and results from the first test day milk as predictors of disease, fertility, yield, and culling in commercial dairy herds. J Dairy Sci 82, 295-304

- Heuer C, van Straalen WM, Schukken YH (2001) Prediction of energy balance in high yielding dairy cows with test-day information. J Dairy Sci 84, 471-481
- Hinrichs D, Stamer E, Junge W, Kalm E (2006) Genetic analysis of several economically important disease traits in German Holstein cows. Arch Tierz 49, 209-221

Jankovská R, Šustová K (2003) Analysis of cow milk by near-infrared spectroscopy. Czech J Food Sci 21, 123-128

- Janů L, Hanuš O, Frelich J, Macek A, Zajíčková I, Genčurová V, Jedelská R (2007) Influences of different milk yields of Holstein cows on milk quality indicators in the Czech Republic. Acta Vet Brno 76, 553-561
- Jílek F, Řehák D, Volek J, Štípková M, Němcová E, Fiedlerová M, Rajmon R, Švestková D (2006) Effect of herd, parity, stage of lactation and milk yield on urea concentration in milk. Czech J Anim Sci 51, 510-517
- Khaled NF, Illek J, Gajdůšek S (1999) Interactions betweens nutrition, blood metabolic profile and milk composition in dairy goats. Acta Vet Brno 68, 253-258
- Koops J, Kerkhof Mogot MF, van Hemert H (1989) Routine testing of farm tank milk by infra-red analysis. IV Prediction of the freezing-point depression from infra-red measurements and conductivity. Neth Milk Dairy J 43, 3-16
- Kubešová M, Fajmon T, Frelich J, Trávníček J, Maršálek M (2009) Analysis of milk urea and milk citrate content during the postpartal period and their impact on reproduction in dairy cows. Výzkum v chovu skotu/Cattle Research Ll, 185, 2-13
- Kukačková O, Čurda L, Jindřich J (2000) Multivariate calibration of raw cow milk using NIR spectroscopy. Czech J Food Sci 18, 1-4
- Kühn C, Reinhardt F, Schwerin M (2008) Marker assisted selection of heifers improved milk somatic cell count compared to selection on conventional pedigree breeding values. Arch Tierz 51, 23-32
- Larson SF, Butler WR, Currie WB (1997) Reduced fertility associated with low progesterone postbreeding and increased milk urea nitrogen in lactating cows. J Dairy Sci 80, 1288-1295
- Meloun M, Militký J (1994) Statistical evaluation of experimental data, 839 [in Czech]
- Miglior F, Sewalem A, Jamrozik J, Lefebvre GM, Moore RK (2006) Analysis of milk urea nitrogen and lactose and their effect on longevity in Canadian dairy cattle. J Dairy Sci 89, 4886-4894
- Mottram T, Masson L (2001) Dumb animals and smart machines: the implication of modern milking systems for integrated management of dairy cows. In: Integrated management system for livestock, occasional Publication 28, British Society of Animal Science, 77-84
- Mottram T, Velasco-Garcia M, Berry P, Richards P, Ghesquiere J, Masson L (2002) Automatic on-line analysis of milk constituents (urea, ketones, enzymes and hormones) using biosensors. Comp Clin Path 11, 50-8
- O'Moore LB (1949) A quantitative diffusion method for the detection of acetone in milk. Vet Rec 31, 457-458
- ON 57 0534: Milk fermentationability determination. Standard Milk Industry Prague 1986 [in Czech]
- Panicke L, Weingärtner J, Schmidt M, Krol T (2000) Relationship between lysosomal blood activity and milk contents of urea and protein in different phases of milk production in dairy cows. Arch Tierz 43, 17-25 [in German]
- Parland McS, Kearney JF, Rath M, Berry DP (2007) Inbreeding effects on milk production, calving performance, fertility, and conformation in Irish Holstein-Friesians. J Dairy Sci 90, 4411-4419
- Rajala-Schultz PJ, Saville WJA, Frazer GS, Wittu TE (2001) Association between milk urea nitrogen and fertility in Ohio dairy cows. J Dairy Sci 84, 482-489
- Reneau JK (1986) Effective use of dairy herd improvement somatic cell counts in mastitis control. J Dairy Sci 69, 1708-1720
- de Roos APW, van den Bijgaart HJCM, Horlyk J, de Jong G (2006) Screening for subclinical ketosis in dairy cattle by Fourier transform infrared spectrometry. Technical presentations, 35th ICAR Session, Kuopio, Finland, June, 53-59
- Řehák D, Rajmon R, Kubešová M, Štípková M, Volek J, Jílek F (2009) Relationships between milk urea and production and fertility traits in Holstein dairy herds in the Czech Republic. Czech J Anim Sci 54, 193-200

- Shook GE (1982) Approaches to summarizing somatic cell count which improve interpretability. In: Nat. Mast. Council, Louisville, Kentucky, 1-17
- Schutz MM, Hansen LB, Steuernagel GR, Reneau JK, Kuck AL (1990) Genetic parameters for somatic cells, protein, and fat in milk of Holsteins. J Dairy Sci 73, 494-502
- Stoop WM, Bovenhuis H, van Arendonk JAM (2007) Genetic parameters for milk urea nitrogen in relation to milk production traits. J Dairy Sci 90, 1981-1986
- Šustová K, Růžičková J, Kuchtík J (2007) Application of FT near spectroscopy for determination of true protein and casein in milk. Czech J Anim Sci 52, 284-291
- Thompson JR, Everet RW, Hammerschmidt NL (2000b) Effects of inbreeding on production and survival in Holsteins. J Dairy Sci 83, 1856-1864
- Thompson JR, Everet RW, Wolfe CW (2000a) Effects of inbreeding on production and survival in Jerseys. J Dairy Sci 83, 2131-2137
- Tsenkova R, Atanassova S, Itoh K, Ozaki Y, Toyoda K (2000) Near infrared spectroscopy for biomonitoring: Cow milk composition measurement in a spectral region from 1,100 to 2,400 nanometers. J Anim Sci 78, 515-522
- Vojtíšek B (1986) The determination of acetone in milk, colostrum, blood and urine of dairy cows by microdiffusion method. Veterinářství 36, 394-396 [in Czech]
- Wood GM, Boettcher DF, Kelton DF, Jansen GB (2004) Phenotypic and genetic influences on test-day measures of acetone concentration in milk. J Dairy Sci 87, 1108-1114
- Xu N, Paul S, Bennewitz J, Reinsch N, Thaller G, Reinhardt F, Kühn C, Schwerin M, Erhardt G, Weimann C, Thomsen H, Mishra S, Kalm E (2006) Confirmation of qualitative trait loci for somatic cell score on bovine chromosome 18 in the German Holstein. Arch Tierz 49, 111-119
- Yazgan K, Makulska J, Weglarz A, Ptak E, Gierdziewicz M (2010) Genetic relationship between milk dry matter and other milk traits in extended lactations of Polish Holstein cows. Czech J Anim Sci 55, 91-104
- Zhai SW, Liu JX, Wu YM, Ye JA, Xu YN (2006) Responses of milk urea nitrogen content to dietary crude protein level and degradability in lactating Holstein dairy cows. Czech J Anim Sci 51, 518-522

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