


Comparison of milk fatty acid profiles measured on Kouri cows near Lake Chad and on dairy cattle as reported by meta-analytical data

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Abstract Kouri (*Bos taurus*) is a breed aboriginal from Lake Chad and threatened with extinction. This study aimed to compare milk fatty acid profiles measured on Kouri cows and on high-yielding dairy cattle in Europe and elsewhere as reported by meta-analytical data (22 experimentations). Milk samples were collected from 14 Kouri dairy cows in dry season (March to June) and fatty acids (FA) were determined by gas chromatography. Overall, 32 FA have been identified. Kouri showed lower values ($P < 0.001$) in the sum of saturated FA (SFA, -10.9 pts), *cis-9, cis-12* 18:2 (-1.00 pt) ($P < 0.01$), higher values ($P < 0.001$) in the sum of monounsaturated FA (MUFA, $+15.3$ pts), C18:0 ($+3.5$ pts), *cis-9, trans-11* C18:2-CLA ($+1.00$ pts), *trans-11* 18:1 ($+1.4$ pts) and ($P < 0.01$) in *cis-9*, C18:1 ($+3.00$ pts) acids. The differences between the milk FA profile of the Kouri cows and that obtained from meta-analytical data could be the possible consequence of the use of particular lake pastures by Kouri cows.

Keywords Kouri breed Lake Chad · Lacustrian pasture · Milk fatty acid · Animal nutrition

Introduction

The Kouri breed (*Bos taurus*) aboriginal from Lake Chad is characterized by some unique adaptation to a lacustrian environment. Raised under natural conditions, age at first calving is about 40 months and average milk production is close to 5 l/day, over about a 333-day lactation (Zeuh et al. 2014; Tellah et al. 2015). The breed actually is endangered owing to frequent hybridization with Zebu breeds (Revue Passages 2015). The composition of FA in milk varies considerably due to differences in genetics and nutrition of cows (Pilarczyk et al. 2015). Thus, the objective of the study was to compare milk fatty acid profiles measured on Kouri cows around Lake Chad with high-yielding dairy cattle in Europe and elsewhere.

Materials and methods

Location of the study area

The study was conducted in the Sahelian zone near Lake Chad (approx. 13° N and 14° E) with 250–500 mm annual rainfall (Revue Passages 2015).

Sampling and chemical analysis

Milk samples (100 ml) were collected from 14 Kouri dairy cows in full lactation, 30% of which were primiparous. The dry matter (DM, method no. 934.01), crude

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Table 1 Brief characteristics of the experimental and meta-analytic data

Authors	Number	Country	Breeds and brief context
Our data	14	Chad	Kouri (<i>Bos Taurus</i>)
Adamska et al. 2014	15	Poland	Polish Red and White; summer
Bardot et al. 2006	20	Pennsylvania	Holstein; High pastures
Bardot et al. 2006	20	Pennsylvania	Holstein; Low pastures
Collomb et al. 2002	21	Switzerland	Simental × Red Holstein; Highland pastures
Collomb et al. 2002	12	Switzerland	Simental × Red Holstein; Mountain pastures
Couvreur et al. 2006	8	France	Holstein
Dohme 2007	6	Switzerland	Brown Swiss and Holstein
Falchero et al. 2010	12	Italy	Italian Red-pie, Alpine grey and Tarantaise; <i>Festuca nigrescens</i>
Falchero et al. 2010	12	Italy	Italian Red-pie, Alpine grey and Tarantaise; <i>Trifolium alpinum</i>
Frelich et al. 2012	4	Czech Republic	Czech Fleckvieh and Holstein
Hurtaud and Delaby 2007	10	France	Holstein
Jahreis et al. 1996	350	Germany	Black-pie
Kay et al. 2005	13	New Zealand	Holstein × Friesian
Khanal et al. 2007	5	Utah (USA)	Holstein
Lock and Gamsworthy (2003)	45	England	Holstein and Friesian
Palladino et al. 2009	68	Ireland	Holstein-Friesian
Palladino et al. 2010	27	Ireland	Holstein-Friesian and Jersey
Rego et al. 2004	12	Portugal	Holstein
Rego et al. 2008	12	Portugal	Holstein
Revello et al. 2010	40	Italy	Aosta red pie
Rouille and Montourcy 2010	55	France	Holstein
Tyagi et al. 2008	5	Pakistan	Crossbreed Holstein-Friesian × Tharparkar

ash (CA, method no. 942.05), and ether extract (EE, method no. 920.39) according to AOAC procedures (AOAC 2006). Crude protein was determined by the Kjeldahl method, with the determination of nitrogen ($N \times 6.39$) for milk (AOAC 2006). The determination of the fatty acid (FA) composition of the fat due milk was made by gas chromatography (GC) after extraction and transesterification. The fatty acid was expressed in percent of total fatty acids (TFA).

Statistical analysis

The fatty acid profiles of the Kouri cows were subjected to standard descriptive statistics (means, standard deviations) and compared using a meta-analytic approach (Cucherat et al. 2002) with those in 22 studies reported in the literature (Table 1).

The published means (M_i) and effective number (n_i) of the groups studied in the literature were recorded. Variances (V_i) were deduced as follows, when supplied with the following: (1) the standard error (SE): $V_i = n_i \text{SE}^2$; (2) the residual standard error (RSE): $V_i = \text{RSE}$; (3) the standard error of the mean (SEM), $V_i = \text{dl}_i \text{SEM}^2$, dl_i representing the degrees of freedom associated with the residual variance; and (4) the standard error of the

difference (SED): $V_i = \text{SED}^{2*} n/2$. Weighted average of literature was then obtained using the following formula:

$$M_L = \frac{\sum_{i=1}^N M_i W_i}{\sum_{i=1}^N W_i}$$

where $W_i = \frac{n_i}{V_i}$. The standard error of the weighted average was considered equivalent to $\text{SE}_L = \sqrt{\frac{1}{\sum_{i=1}^N W_i}}$, where N = the number of publications. The confidence interval was therefore considered equal to $\text{IC}_L = 1.96 \pm \sqrt{\frac{1}{\sum_{i=1}^N W_i}} N$.

The experimental average of the present study (M_e) was compared against the weighted average of the literature using the formula:

$$t = \frac{M_e - M_L}{\frac{n_e * v_e + n_L * v_L}{\sqrt{(n_e + n_L - 2)}} \left[\frac{1}{n_e} + \frac{1}{n_L} \right]}$$

Table 2 Proportions of low level fatty acids not or scarcely reported in the literature and determined in milk fat from Kouri breed near Lake Chad

Fatty acids	Mean (% total FA)	SD
Odd- and branched-chain fatty acids		
<i>Iso</i> C15:0	0.7	±0.2
<i>Iso</i> C16:0	0.7	±0.2
<i>Iso</i> C17:0	0.7	±0.2
C15:0	1.8	±0.4
C17:0	1.04	±0.2
<i>Cis</i> -9 C17:1	0.2	±0.2
<i>Anteiso</i> C15:0	0.8	±0.2
<i>Anteiso</i> C17:0	1.2	±0.5
<i>Anteiso</i> C18:0	0.5	±0.2
ΣOBCFA	7.6	±1.7
Other fatty acids		
C4:0	0.8	±0.1
C6:0	0.8	±0.1
C8:0	0.5	±0.1
C10:0	1.0	±0.4
C20:0	0.6	±0.2
C22:0	0.2	±0.6
<i>cis</i> -9 C14:1	0.6	±0.2
<i>cis</i> -9 C16:1	0.8	±0.3
<i>cis</i> -6, <i>cis</i> -9, <i>cis</i> -12 C18:3 (GLA)	0.2	±0.07
<i>cis</i> -9 C20:1	0.2	±0.08
<i>cis</i> -8, <i>cis</i> -11, <i>cis</i> -14 C20:3 (DGLA)	0.06	±0.02
C20:5 n-3 (⁵ EPA)	0.05	±0.02
<i>cis</i> -7, <i>cis</i> -10 C22:2 (DDA)	0.04	±0.02

SD standard deviation, ΣOBCFA sum odds and branched-chain fatty acids, GLA gamma-linolenic acid, DGLA dihomogamma-linolenic acid, EPA eicosapentaenoic acid, DDA docosadienoic acid

where $V_L = (SE_L)^2 \times n_L \times N$ et $n_L = \frac{\sum_{i=1}^k n_i}{N}$. Significant difference was declared at alpha value of 5% for a value of t corresponding to the Student distribution with $(n_1 + n_2 - 2)$ degree of freedom.

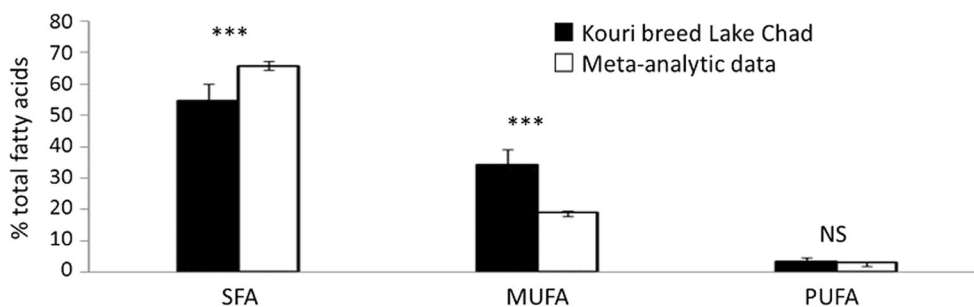


Fig. 1 Proportions of the sum (mean ± standard deviation) of saturated, monounsaturated and polyunsaturated fatty acids in milk fat obtained from Kouri breed near Lake Chad and meta-analytic data from dairy cows as obtained by literature. SFA saturated fatty acids, MUFA

Results

Milk characteristics

Milk DM from Kouri was close to 13%. About one quarter of this was represented by fat (27.8% DM) and another one by protein (26.7% DM). Ash content in DM was close to 6% and lactose content obtained by difference was 39.6%. Consequently, in milk, fat and total nitrogen materials (TNM) were quite similar at 36 g/l while lactose level was higher at 52 g/l. Crude ash was 7.9 g/l.

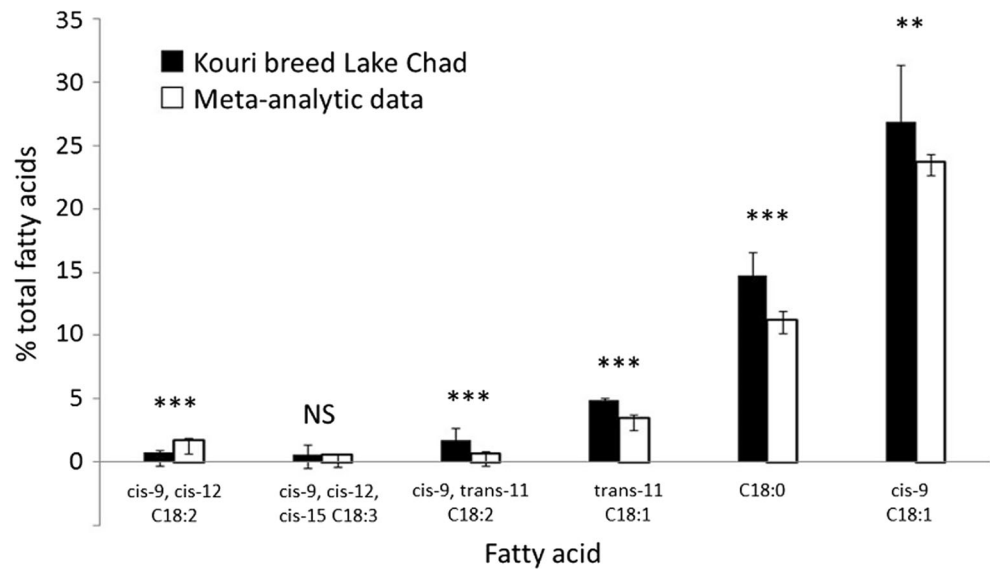
Fatty acid composition of milk

Mean levels and standard deviations for fatty acids obtained in this study but not currently reported in literature, such as odd- and branched-chain fatty acids (OBCFA) and other fatty acid scarcely described, like very short chains from C4 to C10 and long-chain fatty acids (LCFA) are depicted in Table 2. All of them had levels close or lower than 1% of TFA. The highest value was observed for C15:0, at 1.8% TFA. Two LCFA—C20:0 and C22:0—had values lower than 0.8% TFA. Some long-chain fatty acid from C18 to C22, belonging to polyunsaturated fatty acids (PUFA)—*cis*-5, *cis*-8, *cis*-11, *cis*-14, *cis*-17 C20:5, *cis*-7, *cis*-10 C22:2, *cis*-6, *cis*-9, *cis*-12 C18:3, C22:0, and *cis*-8, *cis*-11, *cis*-14 C20:3—showed also values lower than 1% (Table 2).

Concerning FA classically reported in the literature, the proportions of medium-chain carbon fatty acid in TFA were as follows, respectively, in Kouri milk and in data from literature: C12:0, 1.3 ± 0.1 vs $2.9 \pm 0.5\%$; C14:0, 8.00 ± 0.3 vs $10.6 \pm 0.4\%$, % TFA, both at $P < 0.001$; and C16:0, 26.9 ± 0.7 vs $27.8 \pm 0.9\%$, % TFA, NS. A lower average proportion of saturated fatty acids (SFA) (54.8 ± 5.3 vs $65.7 \pm 1.6\%$, % TFA; $P < 0.001$) was obtained in Kouri cows (Fig. 1). By contrast, the sum of monounsaturated fatty acids (MUFA) proportion in TFA was significantly higher in Kouri samples (34.2 ± 1.4 vs $18.9 \pm 0.7\%$, % TFA; $P < 0.001$). It was not the case for the

monounsaturated fatty acids, PUFA polyunsaturated fatty acids; ***($P < 0.001$) very highly significant difference and NS ($P > 0.05$) not significant difference

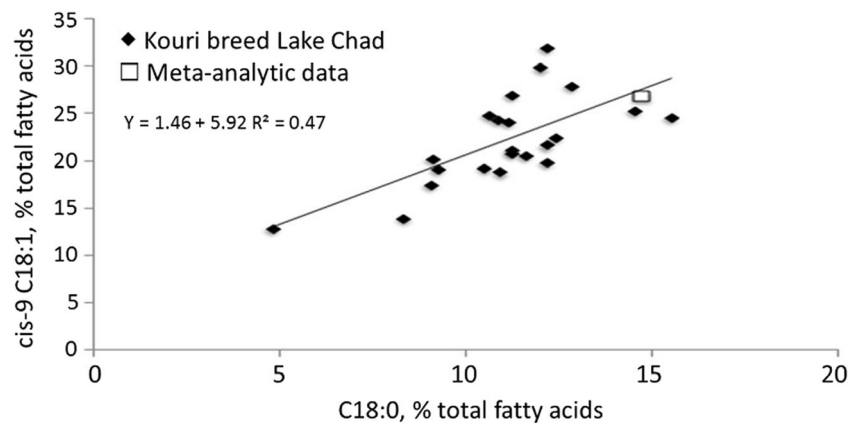
Fig. 2 Proportions (mean \pm standard deviation) of some individual fatty acids in milk fat obtained from Kouri breed near Lake Chad and meta-analytic data from dairy cows as obtained by literature. ***($P < 0.001$) very highly significant difference, highly significant difference **($P < 0.01$) and NS ($P > 0.05$) not significant difference



sum of PUFA (3.4 ± 0.3 vs $3.00 \pm 0.3\%$, % TFA; $P > 0.05$). On the whole in the Kouri breed, the proportions of saturated FA, monounsaturated FA and polyunsaturated FA in TFA were respectively 18% lower, 33% higher and equivalent to average data from the literature.

As reported in Fig. 2, the average concentrations of *cis*-9, *cis*-12 18:2 were low in Kouri breed when compared to literature data (0.7 ± 0.07 vs $1.7 \pm 0.2\%$, % TFA; $P < 0.001$). The mean levels of C18:0 (14.7 ± 0.5 vs $11.2 \pm 0.7\%$, % TFA; $P < 0.001$), *cis*-9, *trans*-11 C18:2 (1.75 ± 0.3 vs $0.7 \pm 0.1\%$, % TFA; $P < 0.001$), *trans*-11 18:1 (4.9 ± 0.04 vs $3.5 \pm 0.2\%$, % TFA; $P < 0.001$) and *cis*-9 C18:1 (27.00 ± 1.2 vs $23.7 \pm 0.8\%$, % TFA; $P < 0.01$) showed very highly significant differences. The relationships between mean oleic and stearic acids values from meta-analytic data and from Kouri are depicted in Fig. 3. The correlation coefficient was 0.68. The values from Kouri fitted into this relationship but were located in the upper levels of the scale. The relationships between *cis*-9, *trans*-11 C18:2 and *trans*-11 18:1 were even stronger ($r = 0.83$), and data from Kouri were also in the upper values (Fig. 4).

Fig. 3 Relationship between C18:0 stearic acid and *cis*-9 C18:1 oleic acid in milk fat obtained from Kouri breed near Lake Chad and meta-analytic data from dairy cow's as obtained by literature



Discussion

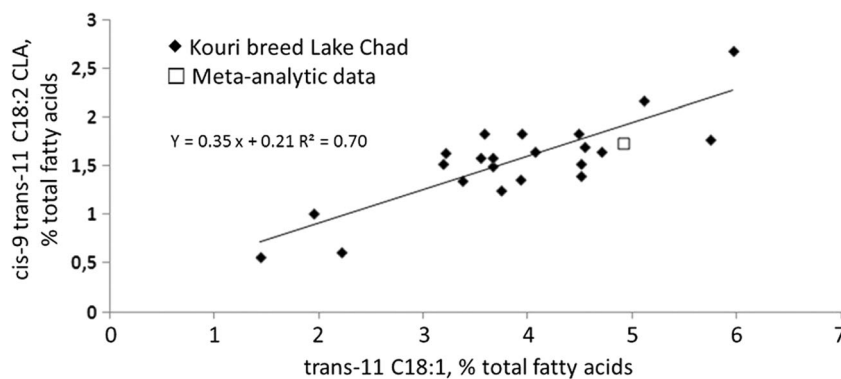
Characteristics of milk

The average milk dry matter in Kouri breed was $133 \text{ g/l} \pm 13$, i.e., between 125 and 135 g/l considered as standard. The fat and protein contents of the milk were 36.8 ± 3.8 and $35.5 \pm 3.5 \text{ g/l}$, respectively. According to Rouille and Montourcy (2010), ranges of 35–45 g/l and 34–35 g/l, respectively, are also considered as normal.

Fatty acid composition of milk

The levels of OBCFA were low but the sum exceeded 13% TFA. The proportion was higher than that obtained from Polish cows in mountain pasture (Adamska et al. 2014). And yet, Falchero et al. (2010) indicated that dairy cows that were grazed on alpine pastures had a milk that was richer in odd-chain saturated FA, such as C15:0 and C17:0 acids. According to Staerfl et al. (2013), the microbial synthesis of

Fig. 4 Relationship between *cis-9 trans-11 CLA* (conjugated linoleic acid) and *TVA* (trans-vaccenic acid) in milk fat from Kouri breed near Lake Chad and meta-analytic data from dairy cows as obtained by literature



branched-chain FA seems to be enhanced by fibre-rich diets. The natural diet of the Kouri breed is probably rich in fibre, owing to the C4 type forages found in tropical countries.

Kouri showed lower values in the sum of SFA—but higher values in C18:0—and linoleic acid (LA) *cis-9*, *cis-12* 18:2, and higher values in the sum of MUFA, *cis-9*, *trans-11* C18:2-CLA, *trans-11* 18:1, and *cis-9*, C18:1, which mainly represents some intermediate forms of saturation of FA, more present with fibre-rich diets (Chilliard et al. 2000). These results confirm those of Ferlay et al. (2013) and Gelé et al. (2014), who reported potentially important differences in fatty acid composition of milk from cows at pasture and cows consuming a total mixed ration in confinement. These variations also are in agreement with potential effects of breed on milk FA composition as reported by Hanuš et al. (2016), comparing Czech Fleckvieh and Holstein breeds. These authors also showed that the proportions of saturated and polyunsaturated FA were lower on summer than on winter grazing, while the monounsaturated and the sum of odd-chain FA contents were higher, as well as those of conjugated linoleic acid (CLA) and α -linolenic acid (C18:3). According to Nantapo et al. (2014), unsaturated fatty acid levels could be improved in milk through pasture feeding and pastures with a high diversity of plants result in higher levels of CLA. Schwendel et al. (2015) indicated that during the grazing season, a leafier plant with less stem provides more unsaturated fatty acids (UFA). According to Glasser et al. (2013), green forages contain 10–30 g/kg DM of total fatty acids composed of 35–70% α -linolenic acid. Roca-Fernandez et al. (2016) reported that differences in milk fatty acids from grazing cows could be related mainly to differences in fatty acid composition of the fodder.

Thus, the differences observed in the present experiment also support some possible effects of grazing on aquatic plant species submerged and floating on lakes, mixed with phytoplankton such as spirulina microalgae

(*Atrhrospira platensis*). Animals swim from island to island to graze aquatic vegetation. The plants preferred by Kouri breed in warm season consist in 60% aquatic vegetation and 35% graminaceae.

Whatever, the Kouri breed appeared to show a strong ability for desaturation by the mammary gland with relatively high concentrations of MUFA, including CLA and low de novo synthesis of fatty acids. The milk from the Kouri breed was richer in *cis-9* C18:1 at about 25% of TFA. This could be explained by a higher level of C18:0 in the milk of this breed. According to Rouille and Montourcy (2010), about 40% of C18:0 collected by the mammary gland are desaturated and contribute to form about 50% of *cis-9* C18:1 secreted in the milk. Similarly, the levels of *trans-11* C18:1 and *cis-9*, *trans-11* C18:2, although low, were higher in Kouri milk. *Trans-11* C18:1 stems from saturation of *cis-9*, *cis-12* C18:2 in the rumen.

Finally, it could be underlined that the milk samples from the Kouri cows studied were especially rich in C18 fatty acids, when compared to the meta-analytical data. The question whether it is an adaptation to lacustrine environment should be further investigated.

Conclusion and outlook

This study underlined some differences in milk fatty acid composition between the Kouri breed and highly productive dairy cows in the world. Milk yielded by Kouri cows reared in lacustrine pasture is a good source of PUFA and MUFA, including vaccenic acid (*trans-11*C18:1) and CLA (*cis-9 trans-11* C18:2), odd- and branched-chain fatty acids. This could be the consequence of the use of particular lacustrine pastures, hence giving rise to new research questions both on the effects of this milk on local human health and on possible metabolic evolutionary adaptations of the breed to its environment. Thus, initiatives including promotion of native dairy cow breeds as a source of valorization should be supported. The

results can be considered as hypothesis generating and may provide relevant scientific justifications for pursuing conservation efforts targeting the Kouri breed.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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