Fatty acids profile, conjugated linoleic acid contents and fat quality in selected dairy products available on the Polish market

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Abstract: The fatty acid composition, *cis-9,trans-*11 C18:2 (CLA) content and lipid quality indices in the fat of some dairy products (pasteurised milk, UHT milk, natural yoghurts, bio-yoghurts, yoghurts with fruit and cereal grains, butters and hard cheeses) available on the Polish market were determined. The conducted study demonstrated that the fat extracted from the analysed dairy products was characterised by various contents of fatty acids and various lipid quality indices. In the fat extracted from all the analysed products, saturated fatty acids (SFA) were dominant. The fat from the yoghurts with the fruit and cereal grains was characterised by the highest content of PUFA (polyunsaturated fatty acids), the highest hypocholesterolaemic/hypercholesterolaemic ratio (H/H) and the lowest value of the index of atherogenicity (AI) and the index of thrombogenicity (TI). The fat from the natural yoghurts contained the highest value of MUFA (monounsaturated fatty acids). The fat from the bio-yoghurts (7.62 mg g⁻¹) had the highest mean content of CLA. In the other analysed products, the mean content of the CLA was significantly lower (P < 0.05), but the fat from the UHT milks (3.32 mg g⁻¹ fat) had the lowest content.

Keywords: dairy products; fatty acids; CLA; lipid quality indices

The consumption of milk and dairy products is frequently included as an important element in a healthy and balanced diet. Milk is the first food for mammals and provides all the necessary energy and nutrients to ensure proper growth and development, being crucial in bone mass formation (Mills et al. 2011). Milk fat contains over 400 different fatty acids, which makes it the most complex of all the natural fats (Jensen 2002). The fatty acid profile of milk fat depends on the fatty acids originating from the feed and the biohydrogenation process that occurs in the rumen. The fatty acid composition depends on many different factors, such as animal species, the breed, season, lactation stage, age, geographical location and, most importantly, the cow's diet (Jensen 2002; Kelsey et al. 2003; Ellis et al. 2006; Frelich et al. 2012; Hanuš et al. 2016). Summer milk contains more unsaturated fatty acids and long chain fatty acids, and less short and middle chain saturated fatty acids than winter milk (Jensen 2002). Not all fatty acids in a specific class have the same effect on human health. The unfavourable feature of milk fat is the high concentration of saturated fatty acids (SFA). These acids increase the LDL (low-density lipoprotein cholesterol) synthesis of cholesterol and increase the risk of coronary heart disease. At present, it is believed that the increased LDL blood concentration is attributable to lauric, myristic and palmitic acids, while the other SFA found in the milk neutralise their effect since they increase the HDL (high-density lipoprotein cholesterol) level (Parodi 2009). In the groups of unsaturated fatty acids, there are acids with high biological activity, such as: oleic, linoleic, linolenic, vaccenic with anticancer and antiatherosclerotic effects (Lim et al. 2014) and conjugated linoleic acid cis-9,trans-11 C18:2 (CLA), which has a number of health benefits, including anticarcino-

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genic, antiatherosclerotic, antioxidative and anti-inflammatory effects (Akalln & Tokusoglu 2003; Aydin 2005; Park 2009; Kee et al. 2010). The aim of the study was to determine the fatty acid composition, the content of the CLA and the lipid quality indices content in the dairy products available on the Polish market.

MATERIAL AND METHODS

Material. The experimental material was: pasteurised milk with 2% fat (12 samples), UHT milk with 2% fat (10 samples), natural yoghurts with from 1.7% to 3% fat (10 samples), bio-yoghurts (natural yoghurts with probiotic cultures, from 1.8% to 3.1% fat) (7 samples), yoghurts with fruit and cereal grains with from 2% to 3% fat (7 samples), butters with 82% fat (12 samples) and ripened rennet cheeses with from 26% to 28% fat (16 samples). The analysed products were from different producers. All the products were bought on the Polish market in the period from May to June 2019. Each sample was analysed in duplicate.

Methods. The fat from the pasteurised and UHT milks was extracted according to the Röse-Gottlieb method (IDF standard 1D:1996, Milk: Determination of fat content – Gravimetric method), the fat from the natural and bio-yoghurts was extracted using Folch's method (Christie 1973). The fat from the yoghurts with fruit and cereal grains and cheeses was extracted according to the Schmidt-Bondzyński-Ratzlaff method (Ładoński & Gospodarek 1986) and the fat from the butters was separated by melting, decantation and filtering (laboratory dryer; Pol-Eko-Aparatura, Poland) through anhydrous sodium sulfate.

The fatty acid methyl esters were prepared according to the IDF method (IDF 182:2002: ISO 15884:2002, Milkfat: Preparation of fatty acid methyl esters). The composition of the fatty acids was determined using the GC-FID (gas chromatography flame ionisation detector) method. The chromatographic separation of the fatty acid methyl esters was carried out on a capillary column (100 m× 0.25 mm i.d. with a film thickness of 0.20 µm) with the CP Sil 88 phase (Varian, USA). The separation conditions were as follows: column temperature from 60 °C (1 min) to °C, $\Delta t = 5$ °C min⁻¹; detector temperature: 250 °C; injector temperature: 225 °C; carrier gas: helium, flow rate: 1.5 mL min⁻¹, and injector: split 50:1. The identification of the fatty acids was carried out based on the comparison of their retention time with the retention time of the methyl esters of the fatty acids of the reference milk fat [Community Bureau of Reference (BCR) material] of the CRM (Certified Reference Material) 164 symbol. The positional *trans* isomers of C18:1 acid were identified using the standards of the methyl esters of these isomers (Sigma-Aldrich, Germany), whereas the *trans* isomers of the C18:2 acid were identified with the use of a mixture of the standards of the C18:2 isomers (Supelco, USA). The *cis-9,trans-11* CLA isomer was identified using a mixture of CLA methyl esters (Sigma-Aldrich, Germany). The contents of the fatty acids were calculated in mg g⁻¹ fat in respect to the introduced standard (methyl ester of the C21:0 acid). The Lipid Quality Indices were calculated according to the fatty acid composition using the following formulae (Ulbricht & Southgate 1991; Osmari et al. 2011):

AI =
$$(C12:0 + (4 \times C14:0) + C16:0) /$$

 $/ (\Sigma n-3PUFA + \Sigma n-6PUFA + \Sigma MUFA)$ (1)

TI =
$$(C14:0 + C16:0+C18:0) /$$

+ $[(0.5 \times C18:1) + (0.5 \times \text{other MUFA}) +$
+ $(0.5 \times \Sigma n-6\text{PUFA}) + (3 \times \Sigma n-3\text{PUFA}) +$
+ $\Sigma n - 3\text{PUFA} / \Sigma n-6\text{PUFA})]$ (2)

$$H/H = (C18:1n-9 + C18:2n-6 + C18:3n-3) /$$
 (3)
 $/ (C12:0 + C14:0 + C16:0)$

where: AI – the index of atherogenicity; TI – the index of thrombogenicity; H/H – the ratio of the hypocholesterolaemic to hypercholesterolaemic fatty acids; PUFA – the polyunsaturated fatty acids; MUFA – the monounsaturated fatty acids; Σn -6 PUFA – the sum of all n-6 PUFA; Σn -3 PUFA – the sum of all n-3 PUFA

Statistical analysis. The statistical analysis was carried out using Statistika 13.1 software. To calculate the significance of the differences, a one-way analysis of variance (ANOVA) was used at a significance level of $\alpha = 0.05$. The differences between the mean values were evaluated with Duncan's test.

RESULTS AND DISCUSSION

The results presented in Table 1 indicate that the fat extracted from the analysed dairy products was characterised by a diversified content of fatty acids. In all the analysed products, saturated fatty acids (SFA) were dominant. The analysed natural yoghurts had the highest fat content (602.72 mg g⁻¹). In the fat from the other analysed products, significantly lower (P < 0.05) contents of these acids were found

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Table 1. The fatty acid composition (mg g⁻¹ fat) and lipid quality indices in the fat from the analysed products

257.23 - 451.29111.43-205.11 39.18 - 77.5211.40 - 20.42.23 - 2.885.37 - 12.790.35 - 0.49 24.83° 1.93^{b} 8.93° $4.88^{\rm b}$ 3.54 - 8.04 3.19^{a} 2.96 - 4.25 3.80^{a} 3.58 - 4.61 179.90^{d} 404.96^{d} 1.97 29.85 165.34-560.80 96.81-236.57 87.23-117.20 22.10-27.56 11.65 - 14.75 $12.96^{b,c}$ $3.12^{a,b}$ 505.78^{b,c} 24.88° 3.59^{b} 2.62 - 4.45 3.79^{b} 2.64 - 5.1491 - 3.483.42 - 3.990.40 - 0.49 0.45° 100.24^{a} 222.00° $3.63^{\rm a}$ Butters 26.48 1.62 0.73 0.99 0.97 11.93 0.21 12 Yoghurts with fruit and cereal grains 178.24-526.46 254.18-273.79 74.82-78.88 19.50-110.61 29.11-98.99 0.89 - 23.05 $504.08^{b,c}$ 4.30 - 32.71.87 - 2.58.84 - 3.040.58 - 0.82 76.83^{a} 2.41^{c} 13.90^{a} 54.95^a 38.33 10.59^{a} 2.20^{d} 261.30^{b} 31.06 16.29 11.33 0.36 0.70^{a} 10.85 0.61 240.27-262.69 Bio- yoghurts 187.38-592.63 56.66-101.09 30.13-39.76 3.41 - 17.420.51 - 0.59 $15.65^{\rm b,c}$ 3.07 - 3.15 258.27^{b} 3.47 - 8.72.82 - 4.86 6.21^{b} $36.08^{\rm b}$ 3.04^{b} .45 - 2.81 0.54^{b} 529.24^{b} 1.65 2.65^{c} 3.12^{b} 5.63 2.50 1.63 0.18 93.87^{a,b} 19.94 86.57–121.00 523.48-658.16 249.35-317.83 6.07 - 29.8229.02-45.51 0.51 - 0.622.98 - 7.103.05 - 7.28..50-2.99 2.92 - 3.50 $38.04^{\rm b}$ 4.79^{b} 21.39^{b} Natural oghurts/ 4.77^{b} 2.66° 0.23 $3.28^{\rm b}$ $0.55^{\rm b}$ 602.72^a 290.37^{a} 27.24 6.11 1.52 5.12 1.65 0.24 10 $\begin{array}{c} 11.18 \\ 62.92 - 103.81 \end{array}$ 111.13-512.14 95.85-227.04 20.08-23.88 10.00 - 12.88**UHT** milks $87.24^{a,b}$ $11.43^{b,c}$ $3.07^{a,b}$ 2.81^{b} 2.33 - 3.06 4.11^{b} 0.43 - 0.462.86 - 3.263.34 - 3.79 22.02^{c} 3.64 - 5.41 3.61^{a} 0.45^{c} 465.39° 207.84° 1.14 0.97 0.62 0.13 0.24 0.11 Pasteurised milks $82.27^{a,b} \\ 22.42 \\ 54.81-111.22$ 89.05-542.38 94.58-244.88 21.34-32.15 1.78 9.23–15.61 $11.42^{b,c}$ $2.86^{\text{b,c}}$ 0.15 3.08 - 5.643.07 - 3.62 225.36° 27.21^{c} 4.04^{b} $2.98^{\rm b}$.95 - 4.370.44 - 0.52.67 - 3.21 $3.28^{\rm b}$ 0.49^{c} 475.19^{c} 44.89 16.66 0.93 0.89 3.72 0.15 nin-max nin-max nin-max nin-max nin-max nin-max nin-max nin-max nin–max nin-max mean mean SD mean mean SD mean mean mean mean mean mean SD SD SD SD SDFatty acids n-6/n-3MUFA SCFA PUFA H/H SFA и-3 9-*u* ΑI Ξ

n – the number of samples; mean – the mean value; SD – the standard deviation; min – the minimum value; max – the maximum value; a, b, c, d – the values denoted in the rows by the different letters indicate statistically significant differences (P < 0.05); SCFA – short-chain fatty acids (C4-C10); SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA - polyunsaturated fatty acids; AI - index of atherogenicity; TI - index of thrombogenicity, H/H - hypocholesterolaemic/hypercholesterolaemic ratio

Significantly, the analysed cheeses had the lowest fat content (404.96 mg g^{-1}).

The SFA group branched-chain fatty acids (BCFA) and odd-chain fatty acids (OCFA) were present in the fat of all the analysed dairy products. The average total BCFA content in the fat extracted from the analysed products ranged from 10.81 mg g⁻¹ of fat in the fat extracted from the cheeses to 22.52 mg g⁻¹ of fat in the fat from the pasteurised milks. The OCFA content was in the range from 12.15 mg g⁻¹ of fat in the fat extracted from the cheeses to 23.67 mg g⁻¹ of fat in the fat from the natural yoghurts. The fat from the butters had the highest content of SCFA (100.24 mg g⁻¹) (Table 1). The fat from the analysed cheeses and yoghurts with the fruit and cereal grains had significantly lower (P < 0.05) contents of these acids. Short-chain fatty acids are important to promote human health (Hanuš et al. 2018).

The analysed natural yoghurts fat (290.37 mg g⁻¹) had the highest contents of the MUFA. Significantly lower (P < 0.05) contents of these acids were in the fat from the other analysed products (Table 1). The study showed that the fat from the yoghurts with the fruit and cereal grains contained the highest content of the PUFA (76.83 mg g⁻¹ of fat). A significantly lower (P < 0.05) content of these acids was in the fat from the other analysed dairy products. The UHT milks fat had the lowest content (22.02 mg g⁻¹) (Table 1).

The study showed that the fats from the yoghurts with the fruit and cereal grains were characterised by a significantly higher (P < 0.05) content of n-3 acids and n-6 PUFA (Table 1). The contents of the n-3 PUFA in the fat from the other analysed dairy products did not differ significantly (P < 0.05). The n-6 PUFA in the fat from the pasteurised milks, UHT milks, natural and bio-yoghurts and butters were on a similar level. The analysed cheeses fat had the lowest content of these acids (8.93 mg g $^{-1}$ of fat).

The n-6/n-3 ratio was the highest in the fat from the analysed yoghurts with the fruit and cereal grains. In the fat from the other analysed dairy products, the n-6/n-3 ratio was significantly lower (P < 0.05), and ranged from 2.98 in the fat from the pasteurised milks to 4.88 in the fat from the analysed cheeses (Table 1). In the raw, pasteurised and UHT bovine milk analysed by Pestana et al. (2015), the n-6/n-3 ratio was 3.51, 3.47 and 3.49, respectively. The n-6/n-3 ratio in the 'Colonia' cheeses produced from cow milk in the autumn was 3.29 and 4.47 in the cheeses produced in the spring (Hirigoyen et al. 2018). According to data in the literature (Wijendran & Hayes 2004; Simopoulos 2008),

an adequate intake of both n-6 and n-3 fatty acids is essential for good health and for lowering the risk of cardiovascular disease and type 2 diabetes. Excessive amounts of n-6 PUFA and a high n-6/n-3 ratio in diets, promote the pathogenesis of many diseases, including cardiovascular disease, cancer, and inflammatory and autoimmune diseases, whereas increased levels of n-3 PUFA (a lower n-6/n-3 ratio), exert suppressive effects (Simopoulos 2008).

In the presented study, the AI value was the highest in the fat from the analysed cheeses (3.19) and the lowest in the fat from the analysed yoghurts with the fruit and cereal grains (2.20). The TI index values were the highest in the fat from the analysed butters and UHT milks (3.63 and 3.61, respectively). Significantly lower (P < 0.05) values were found in the fat from the other analysed dairy products (Table 1).

The lowest TI value (2.41) was found in the fat from the yoghurts with the fruit and cereal grains. According to Ulbricht & Southgate (1991), the AI and TI indices might better characterise the atherogenic and thrombogenic potential of the diet than the PUFA/SFA ratio. The atherogenic index (AI) and thrombogenic index (TI) take the different effects that single fatty acids might have on the human health into account. Thus, the higher the AI, the more the atherogenic dietary components there are. A low AI indicates that the milk and milk products could provide protection against coronary heart diseases. Hirigoyen et al. (2018) report that the AI index and TI index calculated for 'Colonia' cheese produced from cow's milk in autumn and spring were at a similar level, 2.21 and 2,84, respectively.

The H/H ratio is related to the functional activity of the fatty acids in the metabolism of the lipoproteins for the plasma cholesterol transportation and to the risk of cardiovascular disease. Higher values of this ratio are desirable (Santos-Silva et al. 2002). The fat from the analysed pasteurised and UHT milks, butters and cheeses were characterised by similar H/H values (Table 1). A significantly higher (P < 0.05) H/H ratio was found in the fat from the analysed yoghurts. The highest value (0.70) was in the fat from the yoghurts with the fruit and cereal grains.

The *cis-9,trans-*11 C18:2 isomer is the predominant CLA isomer in food. In milk and dairy products, it accounts for more than 80–90% of the total CLA content and is thought to be the isomer, which is characterised by high biological activities (Wahle et al. 2004; Koba & Yanagita 2014). The CLA contribution in the fat from the analysed dairy products is presented in Figure 1. The data shown indicate that the analysed products were char-

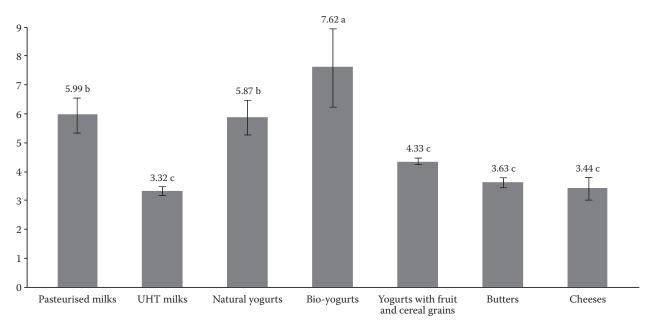


Figure 1. The mean value of cis-9,trans-11 C18:2 (CLA) in the fat from the analysed dairy products (mg g⁻¹ fat)

a, b, c – the values denoted by the different letters indicate statistically significant differences in the content of *cis-* 9, trans-11 C18:2 (CLA) (P < 0.05)

acterised by a different CLA content. According to the data in the literature (Shantha et al. 1995; Lin 2000; Kim & Liu 2002; Sieber et al. 2004; Domagała et al. 2009; Kim et al. 2009), the CLA content ofdairy products is dependent on the initial CLA content of the raw milk, the conditions applied during the technological processes, the additives used and the activity of the starter cultures added. In the analysed products, the fat from the bio-yoghurts (7.62 mg g⁻¹) had the highest mean CLA content.

In the fat from the other analysed dairy products, the mean CLA contents were significantly lower (P < 0.05) (Figure 1). The lowest mean CLA content was found in the fat from the analysed UHT milks (a mean of 3.32 mg g $^{-1}$ of fat). According to Seçkin et al. (2005), the CLA ranged from 1.50 mg g $^{-1}$ to 3.63 mg g $^{-1}$ of fat in Turkish processed cheeses, and from 2.85 mg g $^{-1}$ to 4.67 mg g $^{-1}$ of fat in butter.

CONCLUSION

The conducted study demonstrated that the fat from the analysed dairy products was characterised by various contents of fatty acids, varied contents of conjugated linoleic acid and various values of the lipid quality indices.

The addition of fruit and cereal grains caused a significant increase in the content of polyunsaturated fatty acids in yoghurts. The fat from the bio-yoghurts had the highest mean content of CLA.

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