Composition in fatty acids of mature milk of nursing mothers

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Abstract

Objectives: to determine the fatty acid composition of mature milk of nursing mothers and its distribution according to some maternal variables.

Methods: this is a cross-sectional observational epidemiological study based on the evaluation of the fatty acid profile of mature human milk. Samples of mature breast milk were taken from 106 nursing mothers, by manual milking and who were after the 5th postpartum week. The milk fat extraction was carried out by using the Bligh and Dyer method and methylated with 0.25 mol/L sodium methoxide in methanol diethyl ether. The fatty acid of the milk profile was determined by a Gas Chromatograph equipped with a flame ionization detector.

Results: among the saturated fatty acids, the highest values were observed for palmitic (C16:0), stearic (C18:0), myristic (C14:0) and lauric (C12:0) fatty acids, respectively. Among the monounsaturated fatty acids, there was a higher contribution of oleic (C18:1) and palmitoleic (C16:1) fatty acids, respectively. The total essential fatty acids (linoleic and α -linolenic) was 14.94%.

Conclusions: a low content of essential fatty acids in the breast milk of the nursing mothers was observed in the present study, which are important for infant growth and development. We suggest the need to implement nutrition education strategies aimed for pregnant women and nursing mothers who should be advised to eat healthier foods.

Key words Fatty acids, Human milk, Lactation



Introduction

The role of human milk in child growth and development has been documented for decades. Studies on its composition have allowed to identify various components and their functions, both nutritive and energy substrate supply, as well as in child development.^{1,2}

Lipids are an important source of energy, accounting for more than 40% of the energy intake provided by breast milk for newborns. They also act in the transport of fat-soluble vitamins and as hormonal precursors.³

Additionally, lipids in breast milk play an important role in a child's neurological development, visual acuity, and immune system. The process of development and maturation of these systems occurs markedly in the early years of life and may have repercussions, especially in cognitive aspects, throughout the life of the individual.^{2,4}

The composition of breast milk, especially in relation to its lipid components, undergoes significant variations and is therefore considered a dynamic process that can be modified by factors such as geographic region, duration of breastfeeding, the period of the day, and even during a single feeding. In addition, obstetric factors such as gestational age, 5 anthropometric characteristics 6 and maternal diet 7 have been associated with variations in the lipid profile of breast milk.

The profile and amount of lipids in breast milk varies according to the duration of lactation. Its content in mature milk ranges between 3 and 4 g/dL, approximately 45% to 55% of total caloric value; On the other hand, colostrum has a lower lipid concentration, around 1.8 to 2.9 g/dL, which increases to intermediate values (2.9 to 3.6 g/dL) in transitional milk.8 However, as previously mentioned, this value is modifiable, as intrinsic and extrinsic factors are to the nursing mother, they may influence its fatty acid (FA) composition in human milk.9,10

The growing scientific evidence on the composition of breast milk is an important part of the construction of knowledge about its role in child health and, consequently, its possible consequences in terms of recommendations and public policies. In this context, the present study aimed to determine the composition in fatty acids of mature milk of nursing mothers and its distribution according to some maternal variables.

Methods

This is a cross-sectional observational epidemiolo-

gical study based on the evaluation of the fatty acid profile of mature human milk. The selected population consisted of nursing mothers whose childbirth occurred from August 2014 to December 2015. The sample, chosen sequentially, consisted of 106 nursing mothers, who were the first recruited to the cohort from which this study was originated.

The nursing mothers were recruited from prenatal care in the areas covered by the *Unidades da Estratégia Saúde da Família* (ESF) (Family Health Strategy Units), located in the urban area of Diamantina, Vale do Alto do Jequitinhonha, Minas Gerais, Brazil. It should be noted that the ESF has coverage of approximately 81.5% of the population described.¹¹

Inclusion criteria were: single birth and urban residence. Mothers who had been hospitalized for a long time or were diagnosed with diseases leading to breastfeeding discontinuation were excluded from the study.

A pilot study was conducted to verify if, under real field working conditions, all the proposed logistics would work properly. The study was carried out in a health unit for a period of one week, long enough to achieve the proposed objectives.

Data were collected from a questionnaire that addressed demographic (maternal age), nutritional (pre-gestational BMI and BMI on the third-trimester of gestation) and obstetric history (gestational age) aspects.

Samples (up to 15 mL) of mature breast milk (between 5 and 14 weeks postpartum) were obtained from the same breast offered to the baby by manual milking performed by the woman herself in the morning immediately after the first breastfeeding, before the mother's breakfast, advised by the researchers regarding breast hygiene, the milk collected was in a sterile polyethylene bottle. The milk was transported in a temperature controlled thermal container and stored at -80°C until analysis.

The content of the fatty acid of the milk was determined at the Chromatography Laboratory of the *Universidade Federal de Minas Gerais* (Federal University of Minas Gerais). For this analysis, 0.8 mL aliquots of human milk were used for fat extraction by the Bligh and Dyer method, and 0.25 mol/L methylated sodium methoxide in diethyl ether (1:1).¹²

The analyzes were performed in an HP7820A Gas Chromatograph (Agilent) equipped with a flame ionization detector. EZChrom EliteCompact (Agilent) data acquisition program. A SP2560 30m x 0.25mm x 0.20 ★ m (Supelco) column with temperature gradient was used: 80°C, 0min, 7°C/min up to

240°C; injector (1/30 split) at 250°C and detector at 260°C. Hydrogen as carrier gas (3.0 mL/min) and injection volume of 1 ≒1. Peak identification was made by comparison with FAME C14-C22 methylated fatty acid standards (Supelcocat Number 18917).

The database was entered and stored in the Microsoft Office Excel 2007® and validated in EPI-INFO software, 6.04 version. Data analysis was performed using the Statistical Package for the Social Sciences(SPSS) software, 20.0 version. For statistical analysis, we estimated percentages, means and standard deviations, medians and interquartile ranges of fatty acids present in breast milk, as well as the percentage of minimum and maximum scores.

The normal distribution of the percentage of fatty acids present in the milk was evaluated by the Kolmogorov-Smirnov test. The mean and median percentages of fatty acids according to demographic, nutritional and obstetric variables were evaluated by using: Student's *t* test, ANOVA, Tukey's test and Dunnett's *T3* test for normal variables; and for nonnormal variables, the Kruskal-Wallis test was used.¹³

As this study involved humans, it was submitted to the Ethics Committee and all the precepts of bioethics were carefully followed, obeying the ethical precepts of CNS Resolution Number 466/2012. We were careful to preserve the identity of all study participants. The research project was approved by the Research Ethics Committee of the *Universidade Estadual de Montes Claros* (State University of Montes Claros) under the document number 1,321,802.

Results

Of the 106 nursing mothers evaluated, most were between 20 and 29 years old (48.1%) and had black/mixed skin color (81.2%). Regarding to education, most had between 9 and 11 complete years of schooling (44.3%) and have a paying job (51.5%). When analyzing gestational age at birth, 90.6% of the nursing mothers had children at \geq 37 weeks of gestation. When assessing the nutritional status of the nursing mothers using BMI, it was observed that (68.8%) of the nursing mothers presented adequate weight when evaluating the pre-gestational BMI and (46.8%) presented normal weight when evaluating the BMI by gestational age.

In relation to the fatty acid profile of the nursing mothers' mature milk, saturated, monounsaturated and polyunsaturated fatty acids were identified. The retention time ranged from one minute to seven seconds, where C8:0 (caprylic acid) was identified, and at fourteen minutes, C22:0 (behenic acid) was identified.

Table 1 presents the means and medians of fatty acids present in mature milk of nursing mothers living in the city of Diamantina, Minas Gerais, Brazil. The Kolmogorov-Smirnov test indicates that caprylic acid (C8:0), palmitic acid (C16:0), margaric acid (C17:0), arachidic acid (C20:0), behenic acid (C22:0) and myristoleic acid (C14:1) had nonnormal distribution (p<0.05).

Table 1 also describes the distribution of fatty acids according to the degree of establishment of carbon atoms. Among saturated fatty acids, higher values were observed for palmitic (C16:0), stearic (C18:0), myristic (C14:0) and lauric (C12:0) fatty acids, respectively. Among the monounsaturated fatty acids, there was a higher contribution of oleic (C18:1) and palmitoleic (C16:1) fatty acids, respectively. The amount of essential fatty acids (linoleic and α -linolenic) was 14.94%. In the chromatographic analysis, some unidentified fatty acids were observed. These fatty acids were grouped and named others and corresponded to 2.39% of the fatty acids present in breast milk.

Table 2 presents a comparison between the average percentages of fatty acids in mature breast milk and the maternal age of the nursing mothers. It was observed that according to age groups, the mean percentages of two fatty acids were different (p<0.05). For myristic acid (C14:0), Tukey's test indicated that the age group between 10 and 19 years old showed significantly lower mean compared to the nursing mothers at other age groups.

For linoleic acid (C18:2), the mean percentages in nursing milk according to Dunnett's *T3* test, indicated a statistically significant difference. The group of nursing mothers aged 10 to 19 had higher values than the nursing mothers at other age groups (Table 2).

When the fatty acids were stratified by groups according to the saturation regarding age group, the mean percentages of polyunsaturated fatty acids showed a significant difference between the group of nursing mothers aged 10-19 when compared to others, according to the Dunnett's test (Table 2).

The mean fatty acids in breast milk in relation to the categories of variables such as gestational age at childbirth, pre-gestational BMI and gestational BMI were also compared. However, no significant differences were observed.

For fatty acids that showed a non-normal distribution, the Kruskal-Wallis test was performed to test the differences between the medians of the fatty acid profile in relation to maternal age and gestational

Composition of the fatty acid of breast milk of nursing mothers from Diamantina (MG).

| Fatty acids | RT | $\bar{X} \pm SD$ | Median | Perce | ntiles | Minimum | Maximum | p* |
|----------------------------|-------|------------------|--------|-------|--------|---------|---------|---------|
| | | | | 25 | 75 | | | |
| Saturated | - | 52.19±7.62 | 52.61 | 48.47 | 56.97 | 0.00 | 66.86 | 0.066 |
| C8:0 (caprylic acid) | 1.79 | 0.03±0.14 | 0.00 | 0.00 | 0.00 | 0.00 | 1.43 | < 0.001 |
| C10:0 (capric acid) | 3.08 | 1.03±0.50 | 0.98 | 0.77 | 1.28 | 0.00 | 3.03 | 0.116 |
| C12:0 (lauric acid) | 4.85 | 5.66±2.36 | 5.60 | 4.01 | 6.89 | 0.00 | 11.81 | 0.726 |
| C14:0 (myristic acid) | 6.84 | 8.12±3.08 | 7.87 | 6.21 | 9.77 | 0.00 | 17.16 | 0.464 |
| C15:0 (pentadecanoic acid) | 7.80 | 0.34±0.15 | 0.33 | 0.25 | 0.41 | 0.00 | 1.02 | 0.068 |
| C16:0 (palmitic acid) | 8.90 | 27.18±4.11 | 26.89 | 25.04 | 29.85 | 0.00 | 35.39 | 0.028 |
| C17:0 (margaric acid) | 10.16 | 0.51±0.43 | 0.43 | 0.35 | 0.52 | 0.00 | 3.82 | < 0.001 |
| C18:0 (stearic acid) | 10.76 | 8.17±1.94 | 8.14 | 7.15 | 9.29 | 0.00 | 15.13 | 0.451 |
| C20:0 (arachidic acid) | 12.37 | 0.43±0.75 | 0.28 | 0.20 | 0.42 | 0.00 | 6.91 | < 0.001 |
| C22:0 (behenic acid) | 13.84 | 0.73±0.77 | 0.47 | 0.39 | 0.60 | 0.00 | 4.54 | <0.001 |
| Monounsaturated | - | 31.92±5.25 | 32.10 | 29.18 | 34.46 | 0.00 | 43.02 | 0.230 |
| C14:1 (myristoleic acid) | 7.49 | 0.24±0.22 | 0.21 | 0.12 | 0.31 | 0.00 | 1.33 | 0.029 |
| C16:1 (palmitoleic acid) | 9.72 | 2.25±0.73 | 2.31 | 1.80 | 2.70 | 0.00 | 4.26 | 0.680 |
| C18:1 (oleic acid) | 11.11 | 27.04±4.88 | 26.69 | 24.73 | 29.51 | 0.00 | 38.19 | 0.390 |
| Polyunsaturated | - | 14.94±5.07 | 15.57 | 11.48 | 18.30 | 0.00 | 25.03 | 0.772 |
| C18:2 (linoleic acid) | 11.78 | 13.90±4.74 | 14.41 | 10.72 | 17.01 | 0.00 | 23.52 | 0.726 |
| C18:3 (ɑ-linolenic acid) | 12.58 | 1.04±0.58 | 1.04 | 0.60 | 1.36 | 0.00 | 2.71 | 0.756 |
| Others | - | 2.39±1.07 | 2.39 | 1.77 | 2.85 | 0.00 | 7.53 | 0.090 |

RT = Retention Time; *Komogorov-Sminorv test – if p>0.05 the distribution is normal and the appropriate central tendency measure is the mean.

Fatty acid profile of breast milk and maternal age of nursing mothers from Diamantina (MG).

| Fatty acids | 10 to 19 years old | 20 to 29 years old | 30 or more years old | p* |
|----------------------------|--------------------|--------------------|----------------------|----------|
| | n=20 | n=51 | n=35 | |
| | $\bar{X} \pm SD$ | $\bar{X} \pm SD$ | X ± SD | |
| Saturated | 49.99±5.19 | 53.45±4.58 | 51.62±11.32 | 0.179 |
| C8:0 (caprylic acid) | 0.01±0.03 | 0.02±0.05 | 0.06±0.24 | 0.361 |
| C10:0 (capric acid) | 0.90±0.40 | 1.15±0.51 | 0.93±0.49 | 0.055 |
| C12:0 (lauric acid) | 4.59±1.86 | 6.07±2.29 | 5.66±2.58 | 0.059 |
| C14:0 (myristic acid) | 6.52±2.40 | 8.63±2.88 | 8.29±3.46 | 0.031** |
| C15:0 (pentadecanoic acid) | 0.34±0.14 | 0.34±0.17 | 0.34±0.14 | 1.000 |
| C16:0 (palmitic acid) | 27.10±2.19 | 27.40±3.05 | 26.90±5.97 | 0.856 |
| C17:0 (margaric acid) | 0.47±0.15 | 0.50±0.50 | 0.53±0.43 | 0.870 |
| C18:0 (stearic acid) | 8.87±2.29 | 8.09±1.54 | 7.89±2.21 | 0.178 |
| C20:0 (arachidic acid) | 0.64±1.49 | 0.44±0.53 | 0.31±0.24 | 0.299 |
| C22:0 (behenic acid) | 0.54±0.24 | 0.82±0.92 | 0.70±0.73 | 0.369 |
| Monounsaturated | 32.40±3.48 | 32.51±4.49 | 30.78±6.83 | 0.291 |
| C14:1 (myristoleic acid) | 0.29±0.27 | 0.24±0.25 | 0.21±0.14 | 0.419 |
| C16:1 (palmitoleic acid) | 2.51±0.68 | 2.22±0.64 | 2.13±0.84 | 0.176 |
| C18:1 (oleic acid) | 27.26±2.92 | 27.61±4.57 | 26.10±6.05 | 0.365 |
| Polyunsaturated | 17.60±3.03 | 14.03±5.00 | 14.75±5.65 | 0.026*** |
| C18:2 (linoleic acid) | 16.47±2.73 | 13.07±4.75 | 13.65±5.21 | 0.022*** |
| C18:3 (a-linolenic acid) | 1.13±0.44 | 0.96±0.62 | 1.10±0.60 | 0.430 |
| Others | 2.34±0.59 | 2.44±1.24 | 2.33±1.03 | 0.878 |

^{*}Analysis of variance; ** Tukey's test; *** Dunnett's test.

age at childbirth (Table 3), and pre-gestational and gestational BMI (Table 4) as well. However, no significant differences were also observed.

Discussion

The fatty acid composition of breast milk is variable, and intrinsic and extrinsic factors for the nursing mother contribute to the modulation of these acids. Under these circumstances, the importance of conducting studies to evaluate the lipid composition of human milk and associated factors has been highlighted, since the fatty acids presented in human milk are of great importance due to the peculiarities of their effects on maternal and child health.³

Regarding the results of this study, the highest

Table 3

Fatty acid profile of breast milk and the variable maternal age and gestational age at childbirth of nursing mothers from Diamantina (MG).

| Fatty acids | | Maternal age | | | | | | | |
|--------------------------|--------------------|--------------|--------------------|-------------|----------------------|------------|-------|--|--|
| | 10 to 19 years old | | 20 to 29 years old | | 30 or more years old | | | | |
| | n | =20 | n=51 | | n=35 | | | | |
| | Median | Min-Max | Median | Min-Max | Median | Min-Max | | | |
| Saturated | | | | | | | | | |
| C8:0 (caprylic acid) | 0.00 | 0.00-0.08 | 0.00 | 0.00-0.18 | 0.00 | 0.00-1.43 | 0.707 | | |
| C16:0 (palmitic acid) | 26.93 | 21.96-30.47 | 26.92 | 21.93-33.89 | 26.86 | 0.00-35.39 | 0.964 | | |
| C17:0 (margaric acid) | 0.45 | 0.22-0.93 | 0.43 | 0.00-3.82 | 0.43 | 0.00-2.31 | 0.667 | | |
| C20:0 (arachidic acid) | 0.31 | 0.00-6.91 | 0.28 | 0.00-2.36 | 0.27 | 0.00-1.23 | 0.707 | | |
| C22:0 (behenic acid) | 0.47 | 0.28-1.29 | 0.47 | 0.00-4.54 | 0.47 | 0.00-3.29 | 0.956 | | |
| Monounsaturated | | | | | | | | | |
| C14:1 (myristoleic acid) | 0.27 | 0.00-1.26 | 0.20 | 0.00-1.33 | 0.21 | 0.00-0.50 | 0.367 | | |

| Fatty acids | | Gestational age a | t childbirth | | p* | |
|--------------------------|--------|-------------------|--------------|------------|-------|--|
| | < 37 | weeks | ≥ 3 | ≥ 37 weeks | | |
| | n | =10 | | | | |
| | Median | Min-Max | Median | Min-Max | • | |
| Saturated | | | | | | |
| C8:0 (caprylic acid) | 0.00 | 0.00-0.07 | 0.00 | 0.00-1.43 | 0.325 | |
| C16:0 (palmitic acid) | 28.06 | 21.93-30.75 | 26.78 | 0.00-35.39 | 0.880 | |
| C17:0 (margaric acid) | 0.45 | 0.00-0.66 | 0.43 | 0.00-3.82 | 0.978 | |
| C20:0 (arachidic acid) | 0.16 | 0.00-2.24 | 0.28 | 0.00-6.91 | 0.122 | |
| C22:0 (behenic acid) | 0.46 | 0.00-4.54 | 0.47 | 0.00-3.29 | 0.996 | |
| Monounsaturated | | | | | | |
| C14:1 (myristoleic acid) | 0.16 | 0.00-1.33 | 0.21 | 0.00-1.26 | 0.649 | |
| | | | | | | |

^{*} Kruskal-Wallis's test.

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Table 4

| Fatty acid p | rofile of | breast mill | k and pre-gestation | al and gestational E | BMI of nursing mothers f | rom Diamantina (MG). | |
|--------------|-----------|-------------|---------------------|----------------------|--------------------------|----------------------|--|
| | | | | | | | |

| | | Pi | re-pregnancy BMI (| Classification | | | | | | | | |
|--------|---------------------------------------|---|--|--|--|---|--|--|--|--|--|--|
| Low | veight (n=5) | Overweight (n=14) | | Obesity (n=10) | | Proper weight (n=64) | | | | | | |
| Median | Min-Max | Median | Min-Max | Median | Min-Max | Median | Min-Max | | | | | |
| | | | | | | | | | | | | |
| 0.00 | 0.00-0.09 | 0.00 | 0.00-0.11 | 0.00 | 0.00-0.10 | 0.00 | 0.00-1.43 | 0.836 | | | | |
| 27.08 | 25.51-33.68 | 27.12 | 23.15-31.99 | 27.53 | 22.00-32.99 | 26.68 | 0.00-35.29 | 0.924 | | | | |
| 0.47 | 0.37-0.54 | 0.44 | 0.25-2.31 | 0.45 | 0.25-1.47 | 0.43 | 0.00-3.82 | 0.942 | | | | |
| 0.20 | 0.00-0.35 | 0.32 | 0.00-1.58 | 0.25 | 0.06-1.23 | 0.29 | 0.00-6.91 | 0.274 | | | | |
| 0.47 | 0.29-0.52 | 0.46 | 0.27-0.73 | 0.46 | 0.34-1.27 | 0.46 | 0.00-3.29 | 0.836 | | | | |
| | | | | | | | | | | | | |
| 0.20 | 0.00-0.38 | 0.28 | 0.00-1.33 | 0.18 | 0.00-0.36 | 0.19 | 0.00-0.87 | 0.272 | | | | |
| | 0.00 27.08 0.47 0.20 0.47 | 0.00 0.00-0.09 27.08 25.51-33.68 0.47 0.37-0.54 0.20 0.00-0.35 0.47 0.29-0.52 | Low weight (n=5) Overwee Median Min-Max Median 0.00 0.00-0.09 0.00 27.08 25.51-33.68 27.12 0.47 0.37-0.54 0.44 0.20 0.00-0.35 0.32 0.47 0.29-0.52 0.46 | Low weight (n=5) Overweight (n=14) Median Min-Max Median Min-Max 0.00 0.00-0.09 0.00 0.00-0.11 27.08 25.51-33.68 27.12 23.15-31.99 0.47 0.37-0.54 0.44 0.25-2.31 0.20 0.00-0.35 0.32 0.00-1.58 0.47 0.29-0.52 0.46 0.27-0.73 | Median Min-Max Median Min-Max Median 0.00 0.00-0.09 0.00 0.00-0.11 0.00 27.08 25.51-33.68 27.12 23.15-31.99 27.53 0.47 0.37-0.54 0.44 0.25-2.31 0.45 0.20 0.00-0.35 0.32 0.00-1.58 0.25 0.47 0.29-0.52 0.46 0.27-0.73 0.46 | Low weight (n=5) Overweight (n=14) Obesity (n=10) Median Min-Max Median Min-Max 0.00 0.00-0.09 0.00 0.00-0.11 0.00 0.00-0.10 27.08 25.51-33.68 27.12 23.15-31.99 27.53 22.00-32.99 0.47 0.37-0.54 0.44 0.25-2.31 0.45 0.25-1.47 0.20 0.00-0.35 0.32 0.00-1.58 0.25 0.06-1.23 0.47 0.29-0.52 0.46 0.27-0.73 0.46 0.34-1.27 | Low weight (n=5) Overweight (n=14) Obesity (n=10) Proper weight (n=14) Median Min-Max Median Min-Max Median Min-Max Median 0.00 0.00-0.09 0.00 0.00-0.11 0.00 0.00-0.10 0.00 27.08 25.51-33.68 27.12 23.15-31.99 27.53 22.00-32.99 26.68 0.47 0.37-0.54 0.44 0.25-2.31 0.45 0.25-1.47 0.43 0.20 0.00-0.35 0.32 0.00-1.58 0.25 0.06-1.23 0.29 0.47 0.29-0.52 0.46 0.27-0.73 0.46 0.34-1.27 0.46 | Low weight (n=5) Overweight (n=14) Obesity (n=10) Proper weight (n=64) Median Min-Max Median Min-Max Median Min-Max 0.00 0.00-0.09 0.00 0.00-0.11 0.00 0.00-0.10 0.00 0.00-1.43 27.08 25.51-33.68 27.12 23.15-31.99 27.53 22.00-32.99 26.68 0.00-35.29 0.47 0.37-0.54 0.44 0.25-2.31 0.45 0.25-1.47 0.43 0.00-3.82 0.20 0.00-0.35 0.32 0.00-1.58 0.25 0.06-1.23 0.29 0.00-6.91 0.47 0.29-0.52 0.46 0.27-0.73 0.46 0.34-1.27 0.46 0.00-3.29 | | | | |

| Fatty acids | | | Ge | stational BMI Class | ification | | | | p* |
|--------------------------|--------|---------------|-------------------|---------------------|----------------|---------------|----------------------|---------------|-------|
| | Low v | veight (n=13) | Overweight (n=19) | | Obesity (n=10) | | Proper weight (n=37) | | |
| | Median | Min-Max | Median | Min-Max | Median | Min-Max | Median | Min-Max | |
| Insaturated | | | | | | | | | |
| C8:0 (caprylic acid) | 0.000 | 0.000-0.098 | 0.000 | 0.000-0.163 | 0.000 | 0.000-0.091 | 0.000 | 0.000-1.429 | 0.276 |
| C16:0 (palmitic acid) | 28.865 | 25.626-33.705 | 26.460 | 0.000-35.293 | 25.474 | 23.128-29.886 | 26.576 | 21.930-33.706 | 0.053 |
| C17:0 (margaric acid) | 0.444 | 0.314-0.807 | 0.363 | 0.000-3.818 | 0.426 | 0.252-1.473 | 0.440 | 0.246-0.849 | 0.254 |
| C20:0 (arachidic acid) | 0.339 | 0.195-0.731 | 0.240 | 0.000-0.540 | 0.288 | 0.055-1.234 | 0.308 | 0.000-2.355 | 0.197 |
| C22:0 (behenic acid) | 0.450 | 0.237-1.137 | 0.455 | 0.000-2.267 | 0.538 | 0.368-1.268 | 0.512 | 0.000-3.290 | 0.130 |
| Monounsaturated | | | | | | | | | |
| C14:1 (myristoleic acid) | 0.175 | 0.000-0.528 | 0.256 | 0.000-0.576 | 0.211 | 0.000-0.358 | 0.196 | 0.000-0.545 | 0.560 |
| | | | | | | | | | |

^{*}Kruskal-Wallis's test.

concentrations found were saturated fatty acids, where it was possible to observe higher means for palmitic (C16:0), stearic (C18:0), myristic (C14:0) and lauric (C12:0) acids, respectively. These results were also observed by Silberstein et al.14 who, in a study conducted with Israeli nursing mothers, found similar means of these fatty acids. In studies conducted in Brazil, Nishimura et al.2 and Meneses et al.15 evaluating the fatty acid profile of human milk, found these same fatty acids, but with lower means than the findings of the present study. For the caprylic (C8:0), pamitic (C16:0), margaric (C17:0), arachidic (C20:0) and behenic (C22:0) saturated fatty acids; and myristoleic monounsaturated acid (C14:1), the findings of the present study regarding the concentration of these acids were similar to Nishimura et al.2 and Meneses et al.15

Saturated fatty acids are compounds considered important in the composition of human milk, as they are presented as an energy source or as a substrate for synthesis of intermediate compounds.¹⁵ The production of these fatty acids in human milk is influenced by maternal feeding. Diets composed of low percentage of lipids and high percentage of carbohydrates intensify the production of these acids.¹⁴

Although saturated fatty acids offered to children in the first six months of life through lactation are important in the child's growth and development, the type and especially the amount of these acids that are consumed should be considered.³ The literature reports that lauric acid (C12:0) and myristic acid (C14:0) are potentially cholesterolemic.¹⁶ However, the occurrence of a high palmitic acid (C16:0) content in breast milk ensures greater digestibility, facilitates its use as an energy source, generates other fatty acids or can be stored by the newborn.³ Stearic acid (C18:0), a component that in human tissue is rapidly converted to oleic acid (C18:1), is found at moderate levels related to palmitic acid.¹⁷

Among the monounsaturated fatty acids, there was a higher contribution of oleic (C18:1) and palmitoleic (C16:1) fatty acids, respectively. In Brazil, studies which aimed at assessing the acid profile in breast milk observed similar averages to our findings, both for the total average of monounsaturated fatty acids and for C16:1 and C18:1 acids.^{2,18}

Although they can be synthesized by the human body, the composition and percentage of monounsaturated fatty acids in human milk can be modified by maternal feeding.^{17,19} Due to the many beneficial factors of monounsaturated acids suggested in the literature, the consumption of vegetable oils rich in

these compounds should be stimulated.³ These fatty acids are used by the newborn as an energy source and to make up the membrane structure, with oleic acid (C18:1) being the most commonly type found,^{17,19} corroborating the findings of the present study.

Regarding to the food intake of nursing mothers in Alto do Vale do Jequitinhonha, specifically in the city of Diamantina, Minas Gerais, Brazil, a study by Freitas et al.20 observed a high total intake of vegetables, dark green and orange vegetables, meat, and eggs, oil and saturated fat by nursing mothers. However, despite the consumption of some oleaginous fruit in other regions of a Brazilian savannahlike cerrado, such as pequi (Caryocar brasiliense), which is rich in monounsaturated fatty acids, especially oleic acid,21 the consumption of these fruit was not reported in studies carried out in the city of Diamantina involving nursing mothers²⁰ and pregnant women,²² which may be justified by the seasonality of the fruit,23 or by the data collection instruments used in studies conducted in the city.^{20,22}

Regarding to the means of polyunsaturated fatty acids found in the present study, it was below the averages found in studies conducted in Brazil.^{2,18} Among the polyunsaturated fatty acids, those of the family n-3 and n-6 stand out, as they cannot be synthesized by men. The main acid of the n-3 family is alpha-linolenic acid and of the n-6 family is linoleic acid,¹⁸ the only polyunsaturated acids identified in the present study.

Polyunsaturated fatty acids stand out for their beneficial effects on human health, playing an important role in the development of the central nervous system, retinal cells, and infantile organs.^{2,4} It also has suppressive effects such as inhibition of lymphocyte proliferation, production of antibodies and cytokines, expression of adhesion molecules and Natural Killers (NK) cells activation.²⁴

Studies report the diversity of factors that influence the fat content and FA composition in human milk, among which it is possible to highlight: gestational age at birth,⁵ stage of lactation,⁶ maternal age,⁶ daily variation between lactations,²⁵ genetic factors,²⁶ nutritional status⁶ and eating habits.⁷

When comparing the means and medians of the breast milk fatty acid profile with aspects such as demographic (maternal age), nutritional (pre-gestational BMI and BMI on the third trimester of gestation) and obstetric history (gestational age at child-birth), it was possible to observe a variation in C14: 0 and C18: 2 fatty acid content between the analyzed age groups, in accordance with some studies in which the variable maternal age was found as an

influencing factor of the fatty acid profile of the breast milk.

A study carried out by Azeredo²⁷ observed that adolescent nursing mothers presented a different dietary pattern than adult nursing mothers, presenting higher consumption of goodies, fried foods and processed foods, which are sources of saturated and trans fats, and lower consumption of olive oil, source of monounsaturated fats. Thus, this difference in eating habits between the groups is one of the factors that may explain the different fatty acid profiles found in human milk. The differences observed in human milk can also be attributed to the metabolic differences presented by both groups.

According to Kedem *et al.*,²⁸ advanced maternal age may compromise metabolism, thus affecting milk composition, which suggests that age may be a factor in the fatty acid composition of human milk. However, this information about the possible effect of maternal age on milk fatty acids is still scarce in the literature. Argov-Argaman *et al.*⁶ reports in their study that the fatty acid profile in milk may also be associated with changes induced by maternal dietary fat composition and fat desaturation processes, which may be associated with maternal age. This should be taken into account while planning diets for pregnant women at different ages.

The modulation of FA in human milk is well related to the body composition of the nursing mother.³ Studies have shown that factors such as gestational weight gain or during breastfeeding can determine the concentration of fats (fatty acids) present in nursing mother's milk.^{29,30} However, in the present study, these findings were not confirmed. Thus, studies that seek in knowing about the fatty acid profile of human milk has become important, due to the scarcity of studies in the national literature that associate the profile of milk fatty acids with maternal factors, and these acids are of great importance for maternal and child health.

The main objective of this study was to determine qualitatively and quantitatively the composition of breast milk regarding its fatty acid profile. However, some limitations must be assumed so that

the interpretation of the results shown here is interpreted with caution. Initially, we did not explore all possible factors that could interfere with the profile of fatty acids in breast milk, such as smoking, alcoholism and maternal diet. The interpretation of the results in a cross-sectional study also stands out, given that the composition of breast milk changes depending on lactation time, as well as the fact that milk collection was restricted to only one sample of milk collected from each woman. Thus, it did not cover the variability of fatty acid composition throughout the day.

Few Brazilian studies have evaluated the fatty acid profile of breast milk, which limits the comparison of the results found. Low content of essential fatty acids in the breast milk of nursing mothers of the present study was observed, which are of fundamental importance for the growth and development of the infant. There were significant differences in the means of polyunsaturated fatty acids between the group of nursing mothers aged 10 to 19 and the group of nursing mothers aged 20 to 29. From the fatty acid profile found in the present study, it is suggested the implementation of nutritional education strategies aimed for pregnant women and nursing mothers at different age groups, who should be oriented to consume healthier foods rich in monounsaturated fatty acids (such as olive oil) polyunsaturated (tuna, anchovy, salmon and sardines), reducing the consumption of food high in saturated and trans fats.

Authors' contribution

Freitas RF, Pinto NAVD and Teixeira RA were responsible for the study design and manuscript writing. Macedo MS, Lessa AC, Ferraz VP contributed to the analysis and interpretation of data and revision of the manuscript. Soares NO and Martins BEV contributed to data collection and database construction. All authors have approved the final version of the manuscript and are publicly responsible for the content of the article.

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