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## Fatty Acid Composition of Human Colostrums of Burkinabe Women

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**Abstract:** The aim of this study is to compare/contrast the colostrum lipid composition of 53 Burkinabe women, collected in 2005 at the Maternity of Centre Medical Saint Camille in Ouagadougou (Burkina Faso), with similar data obtained in breast milk, five years ago and then to show the evolution of this important aliment. The fatty acid composition of colostrum samples was determined by Gas-liquid Chromatography-Mass Spectrometry. Saturated lipids (C8:0-C:14:0) showed a progressive increasing trend in the Burkinabe woman colostrum with respect those already measured five years ago. The C15:0-C24:0 fractions were found costantly higher, but their trends were in progressive decrement. The 18:2n-6 fraction (linoleic acid) reached the highest value in the third day post partum. The 18:3n-3 was constantly higher in the second and third days. The 20:4n-6 (arachidonic acid) and LC n-6 PUFA were lower ever since the first day, but with a trend to increase. Also 22: 6n-3 and LC n-3 PUFA were costantly lower. The 18:2n-6/18:3n-3 and LC n-6/LC n-3 ratios were lower and higher, respectively, if compared with those already measured five years ago. These results suggest the need to improve alimentary habits of mothers in order to restore the balanced n-6/n3 PUFA ratio in their colostrums.

**Key words:** n-6 polynsaturated, n-3 polynsaturated, fatty acid, colostrums, Burkina Faso

### INTRODUCTION

A relationship between the alimentary habits of mothers and the concentration of lipid and fatty acids in breast milk has already been reported (Koletzko *et al.*, 2001). These characteristics are more evident in developing countries, where the poor social status, as well as the country characteristics (climate, food security and dietary habits), influence the nutritive composition of breast milk (Rocquelin *et al.*, 1998). In fact a supplement with either LC n-6 or LC n-3 PUFA could be necessary in most developing countries, where the socio-economic status of the population is precarious and dietary habits of mothers do not guarantee advantageous lipid intake necessary for the infant growth. In Burkina Faso Rocquelin *et al.* (2001) have demonstrated that, against an elevated concentration of breast milk lipids  $33.42 \text{ g L}^{-1}$ , the fatty acid composition strongly related to the mother dietary habits. In particular, high portion of linoleic (19.8%), n-6 long chain polyunsaturated (1.90%) and 8:0-14:0 saturated fatty acids (26.94%) have been found.

The diet of these mothers was relatively monotonous, rich in millet flower carbohydrates and poor in fat and proteins. By contrast, an analogous study (Rocquelin *et al.*, 2003) in Brazzaville (Congo), showed that breast milk was rich in n-3 polynsaturated fatty acids (PUFA) (2.39% of total FA) due to the frequent consumption of fish and green leafy vegetables. The milk also contained high 8:0-14:0 FA portions (26% of total FA).

The colostrum lipid composition also appears to be markedly influenced by geographic differences and maternal dietary composition (Fidler and Koletzko, 2000). This, in turn, could influence both growth and development of infants, including cognitive and biological factors since the first week of life.

In this perspective, the aim of the present study is to characterize the lipid content in colostrums of Burkinabe women living in Burkina Faso and to compare results with those obtained in a previous study performed in the same region, in order to demonstrate the effect of social and economical changes on the breast milk lipid composition.

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## MATERIALS AND METHODS

**Study area:** Colostrums of 53 consecutive Burkinabe women were collected between July and October 2005 at the Maternity Centre Medical Saint Camille (CMSC) in Ouagadougou (Burkina Faso - Africa). Burkina Faso (formerly Upper Volta) was once a French colony but gained its independence in 1960. Currently, it is one of the poorest countries in the West Burkinabe region, between Sudan and Guinea. The population (11-12 million) belongs to several ethnic groups (Mossi, Peuhul, Gurunsi, Bobo, etc.). They are primarily shepherds or non-nomadic farmers, living in sod and thatch huts of small and rural villages. Their socio-economic status is poor and their hygienic-sanitary conditions are defective with a bad water supply. The direct consequence of these conditions is that the oro-oral transmission of infectious diseases is easy, from the first day of life.

Ethical approval for the study was received by the local institutional review board (CMSC).

**Subjects:** Data on personal characteristics (age, number of pregnancy, gestational age etc.) and clinical findings (history, symptoms, body temperature, modality of delivery) were collected from all the women participant to the study. Mother food was in accord to the traditional habits of their countries. Millet was the staple food of Burkinabe women. Rice and wheat bread were also frequently consumed. Millet and rice were served with a variety of sauces, including dried green leafy vegetables, sumbala (fermented seeds of Burkinabe locust bean), peanut paste, vegetable oil (cotton, peanut, palm), few times meat or dried/smoked fish and flavoring. Due to the poor Burkina Faso country, the overall caloric intake (aliments and sauces) was also poor. A prevalence of LCn-6 in Burkinabe mothers who consume prevalently vegetables was expected. Anthropometric characteristics, educational and socioeconomic status of mothers are reported in Table 1.

**Milk sample collection:** Mothers for colostrums donation were chosen in order of presentation at the Maternity Department. The exclusion criteria included HIV infection, sexual transmitted diseases and mastitis. All individuals participating in the present study signed informed consent forms before starting the study. Colostrums were collected in the morning before breakfast at 24, 48 and 72 h post partum.

Milk samples were collected using a standardized procedure, in the morning after awakening before breastfeeding their babies. Breast milk were expressed through a standardized procedure with a vacuum pump

Table 1: Anthropometric characteristics, education and socioeconomic status of Burkinabe mothers observed in 2001 and in 2005

	Ouagadougou* (n: 101)	Burkinabe (n: 53)	p-value
Maternal age (years)*	28.6±7.0	26.6±7.0	NS
Height (m)*	1.62±0.05	1.63±0.05	NS
BMI (kg m <sup>2</sup> )*	23.0±4.0	24.0±4.0	NS
Number of Deliveries	4 (1/9)	2 (1/8)	0.0001
Gestational Age (months)	40.0 (39-41)	39.5 (38-40)	NS
Modality of delivery	101/101 spont.	53/53 spont.	
Maternal education (%)			
None	59.4	57.4	NS
Primary school level	23.8	25.9	NS
Secondary school level	17.8	20.7	NS
Higher level	0	1.0	NS
Mother's occupation (%)			
None	64.4	68.8	NS
Homes (%) with			
No electricity	78.2	68.2	NS
No refrigerator or freezer	92.1	82.1	NS
No private water supply	83.2	73.2	NS
No private toilets	97.0	87.0	NS
Radio set	78.2	88.2	NS
TV set	26.7	36.7	NS

BMI-Body Mass Index; NS: Not Significant, \*Values given as mean ± standard deviation or median and range, \*:Rocquelin *et al.* (2001)

and collected into a graduate sterile polystyrene tube, immediately refrigerated at 4°C. Mechanical expression lasted at maximum 10 min and was repeated for three consecutive days at 24 h intervals. After the collection, all the mothers continued to breastfeed *ad libitum* their babies at CMSC for three days before going at home. The volume of colostrum was measured and the samples were fractioned in other polystyrene tubes of 2 mL. They were transported on ice to the local laboratory and stored at -20°C. All samples were subsequently sent in dry ice to the Institute of Food Sciences, CNR, Avellino, Italy for the analysis of lipid composition.

**Lipid analysis:** The colostrums samples were centrifuged at 680×g at 4°C for 10 min and the liquid component was removed and saved for other studies. The lipid stratus was manually collected in Eppendorf vials and the total lipid content was gravimetrically measured. Fatty acid methyl ester analyses were performed by using a GCT™ orthogonal acceleration-Time-Of-Flight (oa-TOF) GC mass spectrometer (Micromass Waters Corporation, Atlas Park, Manchester, UK) equipped with a gas chromatograph GC System 6890 Series (Agilent Technologies, Palo Alto, CA, USA) fitted with a capillary column (30 m×0.25 mm internal diam., 0.25 µm film thickness) DB-1 (J and W Scientific, Folsom, CA, USA). Mass spectrometric analyses were performed at 70 eV ionization energy; the 50-500 mass unit range was analysed with a scan time of 0.5 sec. The relative amounts of fatty acids were achieved from their peak areas.

**Statistical analysis:** The statistical analyses were based on the assumption of the normal distribution of variables. All data are expressed as mean±standard deviation (SD). For individual fatty acid and total fatty acid categories mean and SD were calculated for the observation number (n) of each group. Differences among the means were compared using T test for non paired data and a level of p<0.05 was considered significant. The chi-square was used to compare qualitative variable presented as percentages (maternal education and socioeconomic status, feedings practices and health status and modalities of delivery). The multiple linear correlation analysis was calculated with SPSS-10 program for Windows (SPSS Inc, Chicago, Illinois, USA).

### RESULTS

The antropometric characteristics, education and socioeconomic parameters of Burkinabe mothers, collected in 2005, showed a slight improvement, when compared with the same parameters of five years ago. However their social levels remained low in general and did not reach a statistical significant difference. The number of pregnancies was higher in Burkinabe women. (Table 1).

The colostrum lipid content and PUFA profiles of 53 Burkinabe women are reported in Table 2 and compared with similar parameters obtained by Rocquelin *et al.* (2001). It results evident that the total lipid concentration was increasing in the three days (from 2.9±1.2 to 4.3±1.7 g dL<sup>-1</sup>) for the Burkinabe colostrums, comparable to total lipid concentration of Ouagadougou women (3.34 g dL<sup>-1</sup>). Saturated lipids (C8:0-C:14:0) showed a progressive increasing trend and were lower

than before. The C15:0-C24:0 fractions were constantly higher with a progressive decreasing trend. Monounsaturated fractions were higher with a slight tendence to decrease. The 18:2n-6 fraction that corresponds to the linoleic acid, was lower. The 20:4n-6 that corresponds to arachidonic acid, was lower but with a progressive increment. LCn-6 PUFA was lower but with a trend to increase. The 18:3n-3 fraction (linolenic acid) was constantly higher. Both 22:6n-3 and LCn-3 PUFA fractions were constantly lower even though the latter showed an increasing trend.

The 18:2n-6/18:3n-3 ratio was always lower (between 25:1 and 47:1) compared to data of Rocquelin *et al.* (2001) with a progressive decrement during the first three days. The LCn-6/LCn-3 ratio was constantly higher (between 11:1 and 15:1) and showed a decrement because of the increment of LCn-3. A significant correlation between 20:4n-6 (arachidonic acid) and 22:6n-3 (DHA) was observed only in the first two days (Fig. 1). A significant correlation was found between 20:4n-6 and LCn-6 PUFA and between 22:6n-3 and LCn-3 PUFA in Burkinabe colostrums during the first three days. No significant correlations were found between 18:2n-6 and 20:4n-6 nor between 18:3n-3 and 22:6n-3. Correlations between 18:2n-6 and LC-6 PUFA and between 18:3n-3 and LC -3 PUFA were also not significant.

### DISCUSSION

Changes in social and economic status could correspond to improved characteristic of lipid composition in colostrum of Burkinabe women. In fact differences in lipid composition were evident in the colostrums of Burkinabe women collected in 2005, if compared to the breast milk collected in 2001 especially in

Table 2: Fatty acid composition (% w/wt) in 53 Burkinabe colostrums, compared to the same parameters reported by Rocquelin *et al.* (2001) in 101 Ouagadougou breast milk

	Ouagadougou (breast milk)* (n: 101)	p-value <sup>+</sup>	Burkinabe (colostrum) 1° day (n: 53)	p-value <sup>^</sup>	Burkinabe (colostrum) 2° day (n: 53)	p-value <sup>o</sup>	Burkinabe (colostrum) 3° day (n: 53)
TotalLipid content (g dL <sup>-1</sup> )	3.34±0.98	0.001	2.9±1.2	0.019	3.5±1.4	0.009	4.3±1.7
Saturates							
C8:0-C14:0	26.73±10.44	0.001	11.15±3.18	0.001	14.29±3.27	0.022	14.90±3.45
C15:0C24:0	24.76±8.05	0.001	39.56±3.98	NS	38.79±3.50	0.001	34.80±3.92
Monounsaturates	24.61±8.62	0.001	34.35±4.49	0.003	31.91±3.58	NS	32.72±4.07
PUFA							
18:2n-6	20.15±8.48	0.001	13.48±5.73	NS	12.96±3.02	0.002	15.02±3.55
20:4n-6	0.63±0.23	0.001	0.17±0.10	0.001	0.26±0.08	0.025	0.30±0.10
LC n-6 PUFA	1.88±0.68	0.001	0.87±0.40	0.005	1.07±0.32	0.001	1.21±0.34
18:3n-3	0.45±0.24	0.019	0.29±0.02	0.001	0.57±0.18	NS	0.53±0.07
22:6n-3	0.24±0.15	0.001	0.06±0.03	0.001	0.06±0.03	0.001	0.08±0.03
LC n-3 PUFA	0.45±0.26	0.001	0.07±0.05	0.014	0.09±0.03	0.001	0.12±0.04
Ratios							
18:2n-6/18:3n-3	52.6±27.6		47.30±28.02	0.001	25.71±7.97	0.001	30.36±5.34
LC n-6/LC n-3	4.9±1.9	0.001	15.09±8.64	NS	13.00±4.56	NS	11.51±4.48

LC: long-chain; PUFA: polyunsaturated fatty acids, \*: Rocquelin *et al.* (2001); NS: Not Significant, +: Ouagadougou VS Burkinabe (3° day), ^: Burkinabe (1° day) VS Burkinabe (2° day), °: Burkinabe (2° day) VS Burkinabe (3° day)

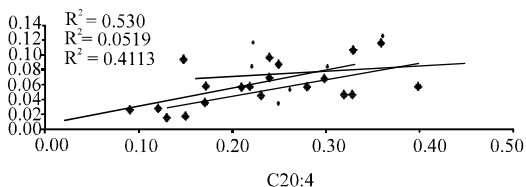


Fig. 1: Correlation between C20:4 and C22:6 in Burkinabe women

the first days. One could object that the colostrum lipid composition could change in the first milk than the last milk of the same breast, but we collected the colostrums in the same time (10 min) in three consecutive days. The difference is evident for monounsaturated and PUFA, consequently, the 18:2n-6/18:3n-3 and LCn-6/LCn-3 ratios are respectively lower and higher in the colostrums of Burkinabe women. In our study, the lack of correlation between essential fatty acids (18:2n-6 and 18:3n-3) and their long chain metabolites (LCn-6 PUFA and LCn-3 PUFA) suggest that the diet availability of essential fatty acids can influence the fatty acid composition of human colostrums. In fact, as already observed by Rocquelin *et al.* (2001), the lipid composition of mature milk of Burkina Faso women was characterized by an elevated content of C8:0-C14:0 because of the mother's diets were rich in carbohydrates and poor in fat. This enhances the biosynthesis of C8:0-C14:0 in the mammary gland during the lactation. Moreover, the 18:2n-6 and LC n-6 PUFA portions were also large because of the typical cereal rich alimentation that contains high quantity of linoleic acid oils (cotton and peanut). In this study the occurrence of high portions of LCn-6 PUFA found in mature milk of Burkina Faso women (Rocquelin *et al.*, 2001) could also be due to enhanced metabolic conversion of 20:4n-6 rather than to dietary sources of LCn-6 PUFA (meat and fish) which, on the contrary, are scarcely available. In fact, the correlation between 20:4n-6 and LC n-6 PUFA, we found in colostrums, is largely expected, thus confirming the metabolic relationship among these two lipid fractions. On the contrary, the high n-3 PUFA level in mature milk (Rocquelin *et al.*, 2001) could be due to dietary intake of these FA (leafy green vegetables). Moreover, the inhibition of the metabolic 22:6n3 to n-3 PUFA conversion, determined by high 18:2n-6 intake, is excluded since the correlation between 22:6n-3 and LC n-3-PUFA was high as expected. This would imply that with advancing lactation, milk PUFAs provision sources gradually shift from adipose tissue catabolism to maternal diet which clearly has improved in this last years.

In addition, the lower concentration of arachidonic acid observed in the colostrum of Burkinabe women, which correlates positively with DHA during the first two days, is not a consequence of any metabolic unbalance. On the contrary, it could be a characteristic of Burkinabe women which exert a protective mechanism on their children from the inflammatory reactions, since arachidonic acid is a prostaglandine precursor. Moreover, an assumption of low quantities of arachidonic acid within breast milk could accelerate the Botallo duct closure in Burkinabe infants, with an undoubt advantage for the newborns (Seidner *et al.*, 2001; Richard *et al.*, 2004). Also the DHA low concentration in the Burkinabe woman colostrums could represent an indirect advantage for Burkinabe newborns. There are several demonstrations that DHA in mothers prevents the postpartum depression (Hibbeln, 2002; Otto *et al.*, 2003). Therefore, in countries where mothers eat low quantities of leafy green, vegetable and fish, the poor DHA secretion in breast milk could be designed in order to preserve this important lipid for the mother, especially in the postpartum period.

In any case, the significant correlations between 20:4n-6 and LCn-6 PUFA and between 22:6n-3 and LCn-3 PUFA in Burkinabe colostrums during the first three days mean that the PUFA synthesis is efficient to ensure the availability of LCn-6 and n-3 PUFA, important for the brain development (Immis, 2000; Jonsbo *et al.*, 1995; SanGiovanni *et al.*, 2000). On the contrary, the absence of a significant correlation between 18:2n-6 and 20:4n-6 and between 18:3n-3 and 22:6n-3 demonstrates that 18:2n-6 (linoleic acid) and 18:3n 3 (linolenic acid) concentrations are always diet-dependent during the first weeks of lactation, as was demonstrated in Burkinabe mothers (Rocquelin *et al.*, 1998).

In conclusion the differences between the actual Burkinabe colostrum composition and that already studied by Rocquelin *et al.* (2001), demonstrate that the diet lipid composition influences the lipid composition of breast milk that, in turn, improves according to the social and economical status of Burkina Faso. These results suggest the need to improve alimentary habits of mothers in order to restore the balanced n-6/n3 PUFA ratio in their colostrums.

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