

Original Research Article

Evaluation of Nutrient Composition in Breast Milk of Breast feeding Mothers in Urban and Sub-urban Subjects in Rivers State

Abstract

The human breast milk is considered to be the perfect food for infants, specifically adapted to their needs and changes in lifestyle and environment may impact on breast milk composition. The aim of this study was to compare the nutrients composition in breast milk in postpartum women in urban and sub-urban areas in Port Harcourt. The study was conducted among 59 postpartum subjects between 0 and 10 days of child delivery in each group. Sampling was done through a simple randomized system. Human breast milk was collected by means of a manual breast pump for the analysis of carbohydrate, protein and lipid using ClegAnthrone Method, Kjeldahl Method and Soxhelt Extraction Method respectively. The results revealed that carbohydrate level was $6.1 \pm 0.5\%$ in urban group and $5.0 \pm 0.1\%$ in sub-urban group which was statistically significant (t-value=2.2; p-value=0.04). Protein level was $3.5 \pm 0.6\%$ in urban group and $3.9 \pm 0.3\%$ in sub-urban group which was statistically non-significant (t-value=0.6; p-value=0.57). Lipid level was $6.1 \pm 0.5\%$ and $5.0 \pm 0.1\%$ in urban and sub-urban groups respectively which was statistically non-significant (t-value=1.2; p-value=0.27). This study has revealed that differences in settlements (urban and sub-urban) have no impact on breast milk composition except in carbohydrate.

Keywords: breast milk, carbohydrate, protein, fat, urban, sub-urban

1.0 Introduction

Human breast milk is produced by human mammary gland and serves the primary role of nourishment for the newly born baby. As discussed in a review by Mead and his team, breastfeeding provides the infant with a number of nutrients and substances that protect and facilitate the growth of the brain including selenium, glutathione, vitamin E, cysteine, tryptophan, choline, taurine, S100B protein, sialic acid, and polyunsaturated fatty acids [1]. The breast milk serves as the primary food for babies until they are mature enough to take solid food, therefore, the quality of nutrient composition is very important for the growth and general health

of the baby, which is why the evaluation of the nutrient composition of breast is necessary. According to World Health Organization (WHO), breastfeeding has health advantages for both the mother and the kid [2]. The content of human milk has been found to be influenced by race, environment and lifestyle [3]. Another research underlined the importance of ethnicity and showed the difference in the amounts of human milk vitamin D [4]. It was discovered that the high levels of osteopontin in breast milk in Asian women are more frequent than in Danes, according to ethnic groups [5]. Therefore, changes in environment and living conditions could be considered as background contributors to nutrient composition in breast milk. Several studies have been conducted on evaluation of nutrient composition in breast milk in other parts of the world but assessment of nutritional composition of breast milk in poorly developed areas like in sub-urban and rural settlements have been poorly studied. Little or no work of such has been conducted in this part of the country (Port Harcourt). The aim of this study was to compare the nutrient composition in breast milk in postpartum women in urban and sub-urban areas in Port Harcourt.

2.0 Materials and Methods

2.1 Study Area

The research study was conducted at clinics and hospitals in Port Harcourt and Eleme. Port Harcourt is the capital city of Rivers state and it is located in the south-south geopolitical zone of Nigeria.

Comment [MP1]: In Which clinic/hospital you conducted your study? Please clearly mention the clinic or hospital where you conducted your study .

➤ Study design is missed. Please describe your study design clearly.

2.2 Study Population

The study was conducted among postpartum subjects between 0 and 10 days of child delivery. The subjects were recruited at the Postnatal Clinic.

Comment [MP2]: It would be fine, if you add source population.

Inclusion Criteria

The following are the inclusion criteria

- Subjects will be registered with the clinic or hospital
- Subjects between 0 and 10 days of childbirth
- Subjects attending postnatal clinic
- Subjects between the ages of 18 and 45

Comment [MP3]: Please use past tense

Comment [MP4]: This would not be an inclusion criteria because your study population already from postnatal clinic

- Subjects must reside within Port Harcourt and Eleme

Exclusion Criteria

The following are the exclusion criteria:

- Subjects not registered and managed by the clinic or hospital
- Subjects with impaired breast milk production

➤ **Sampe size is missed, it would be fine if you describe how you calculate the sample size that in line with the scientific method.**

2.3 Sampling Method

All subjects who met the eligibility requirements and provided their written consent were recruited for the study. Port Harcourt is a metropolis and subjects recruited from this area were categorized as “urban group” while subjects recruited from Eleme area were categorized as “suburban group”. In a simple randomized method, samples were collected from subjects. Subjects were asked to choose from a container having a numbering system of “0” and “1” and all subjects who picked “1” were selected and those who picked “0” were not selected.[6,7]

.....

If it make sense for you ,I rewrite here below:

Sampling technique

Clients/Patients attending the postnatal clinic/hospital were selected by simple random sampling technique.. The list of patients was obtained from hospital registry and the adequate numbers of study subjects was recruited at each clinic /hospital. Selection of study subjects was done using simple random sampling technique. Subjects were asked to choose from a container having a numbering system of “0” and “1” and all subjects who picked “1” were selected and those who picked “0” were not selected

Comment [MP5]: What is simple randomized method? Better to said simple random sampling method

2.4 Specimen Collection

In this study, the manual breast pump technique or method was used in collecting breast milk from postpartum women[8]. In the collection room, the subjects were asked to partly undress in a manner that the breast was revealed and then the pump was applied to drain breast. After collection, the milk was transferred to an appropriate container for storage or immediate laboratory analysis.

2.5 Sample Analysis

Carbohydrates

BY CLEG ANTHRONE METHOD

A 25ml volumetrical flask has been weighed by 0,1g sample and a 62% perchloric acid by adding 1ml distilled water and 1.3ml, shaking for the whole 20 minutes to homogenize the sample. The bottle has been produced using distilled water and a stopper up to 25 ml mark. The solution was filtered or allowed to settle and decant through glass filter paper. 1 ml of filtrate has been collected and placed in a 10 ml test tube, it has been water distilled to volume. The pipette was put to a clean test tube with 1 ml of solution and 5 ml of anthrone reagent. Similarly, 1ml of distilled water was combined with 5 ML of anthrone reagent and the whole blend with 1ml of distilled water and 5ml of anthrone reagent read in blank form at 630nm of the wavelength. Lactose solution 0.1 ml has also been developed and treated with anthrone reagent as a sample. The standard lactose absorption was read and the carbohydrate value was calculated using the following form.

Calculation;

$$\% \text{CHO as lactose} = \frac{25 \times \text{absorbance of sample}}{\text{Absorbance of standard lactose} \times 1}$$

Nutrients Analysis (Protein) By Kjeldahl Method

Stage 1: Digestion

The sample weighed 0.1g into a clean conical bottle of 250ml capacity, put 3g of digestive catalyst to the bottle and also added 20ml sulphuric acid, and heated for digestion. Content of the sky colouring from black to blue. The digest was refrigerated to room temperature with distilled water diluted to 100ml.

Stage 2: Distillation

Diluting the 20ml of a digest into a distillation fibre was measured and the flask was stored on the electrostatic or hot plate heater. A slim condenser linked to a receiver holding 2% boric acid indicator was attached to the distillation flask. A syringe connected to the steel head of the single-arm steel injects 40ml of sodium hydroxide into the digest till a strong digestive alkaline develops. The mixture was brought to boil and the ammonia gas was distilled into the beaker via the condenser. The colour, when the ammonia distillate was added into boric acid, changed from purple to greenish.

Stage 3: Titration

The distillate has been titrated from greenish using the standard solutions for hydrochloric acid 0.1N. This change was recorded as a titre value by the volume of hydrochloric acid added.

Calculation;

$$\% \text{ crude protein} = \frac{\text{titer value} \times 1.4 \times 100 \times 100}{1000 \times 20 \times 0.1}$$

Where;

Titer value = volume of HCl used in titrating the ammonium distillate

1.4 = nitrogen equivalent to the normality of HCl used in the titration 0.1N

100 = the total volume of digest dilution

100 = percentage factor

1000 = conversion factor from gram to milligram

20 = integral volume of digests analysed or distilled

0.1 = the weight of sample in gram digested

Lipid

By Soxhelt Extraction Method

Procedure

In a filter paper, 2g of the material was placed in an extractor soxhlet. A weighted dry distillation flask was put in the extractor. Then the distillation flask was inserted through the condenser end connected to the extractor of the soxhlet (acetone). The installation was conducted using a stand clamp. Cooled water jet could flow into the condenser, which resulted in the refluxation of the heated solvent. The lipid was removed via continuous refluxation in the solvent chamber. The flask was then fully dried in the air oven and weighed again, to get the weight of the lipid, when the lipid was removed from the test sample, the condenser, the extractor and the solvent were detached and evaporated for lipid concentration.

Calculation;

$$\% \text{ lipid} = \frac{\text{Weight of flask and extract of empty flask} - \text{weight of empty flask}}{\text{Weight of sample extracted}} \times 100$$

2.6 Statistical Analysis

Data collected were recorded in Microsoft Excel spreadsheet and were analyzed using SPSS 21.0. Descriptive statistics was done, such as the mean and standard deviation to determine the central tendency and the measure of spread of each variable. T-test was also done to determine if there was a significant difference in the means of the groups (urban group and sub-urban group). The level of statistical significance was set at $p < 0.05$.

3.0 Results

Table:1: Comparing the mean levels of carbohydrate, protein and lipid

Comment [MP6]: T- test is used for small sample size(N<30),if your sample is more than 30 ,it is better use regression analysis

N = 59

SS = statistical significant

In comparing macronutrients (carbohydrate, protein and lipid), two groups were considered (urban and sub-urban groups) as shown in Table 2. In urban group, carbohydrate level was

<i>nutrients</i>	<i>Urban</i>	<i>Sub-urban</i>	<i>T-value</i>	<i>P-value</i>	<i>Remark</i>	
Carbohydrate (%)	6.1±0.5	5.0±0.1	2.2	0.04	Ss	6.1±0.5
Protein (%)	3.5±0.6	3.9±0.3	0.6	0.57	Ns	% and
Lipid (%)	2.4±0.3	3.8±1.1	1.2	0.27	Ns	5.0±0.1

group (T-value = 2.2; P-value <0.05). In urban group, protein level was 3.5±0.6% and 3.9±0.3% in sub-urban group (T-value = 0.6; P-value >0.05). In urban group, lipid level was 2.4±0.3% and 3.8±1.1% in sub-urban group (T-value = 1.2; P-value >0.05).

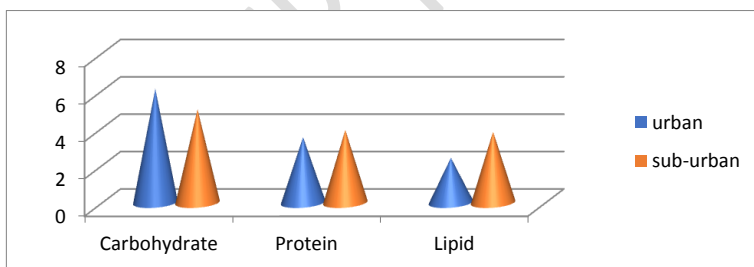


Fig 1: Showing chart presentation of carbohydrate, protein and lipid

4.0 Discussion

In this study, two groups were considered within a postpartum period of 10 days; the urban group had 59 participants and sub-urban group also had 59 participants of postpartum women with mean age of 35±5.0 yrs and 28±3.0 yrs respectively. Their breast milk samples were assayed to determine the variation or changes in nutrient composition between the two groups. Studies have

reported that nutrient composition varies throughout the lactation period and over a single meal [9]. In this study on macronutrient composition in breast milk of lactating mothers, it was found that the carbohydrate level was higher in urban group than in the sub-urban group. The mean difference between the two groups was statistically significant. This implies that the breast milk of postpartum women in the urban area had higher percentage of carbohydrates ($6.1 \pm 0.5\%$). This difference between carbohydrate levels in both groups could be due to changes in lifestyle and feeding habits between both groups. Among all the nutrients assayed, carbohydrates was higher in percentage than others because in early breast milk, studies have reported that carbohydrate provides the main source of energy for the first 12 months, and as such the level of carbohydrate remains higher than other nutrients but declines after 12 months and lipid replaces it as the energy source. This study is in consonance with other studies pointing that carbohydrate has the highest level among other nutrient within the first 12 month [10]. This pattern was consistent in both groups (urban and sub-urban) studied: in the urban group, carbohydrate had the highest level ($6.1 \pm 0.5\%$); protein ($3.5 \pm 0.6\%$) and lipid ($2.4 \pm 0.3\%$). Also in the sub-urban group carbohydrate had the highest level ($5.0 \pm 0.1\%$), protein ($3.9 \pm 0.3\%$) and lipid ($2.8 \pm 1.1\%$). The mean difference in the protein levels between both groups was not statistically significant. The same applies to lipid; the difference in mean levels between both groups was not statistically significant.

Conclusion

This work has shown that nutrient composition in breast milk of postpartum women may not vary based on urban and sub-urban settlements except carbohydrate but changes may be significant in extreme settlements such as urban and rural but that was not within the focus of this study. Further studies may be required to look at changes in nutrients composition in urban and rural areas.

Recommendation

.....

Acknowledgment

.....

Comment [MP7]: Conclusion is the main finding of your work. So that it is depend solely on your findings, however the things which I marked here should be written under recommendation section.

References

1. Mead, M. N. (2008). Contaminants in human milk: weighing the risks against the benefits of breastfeeding. *Eviron Health Perspect*, 116(10), 4427-4434
2. WHO. Preventing disease through healthy environments exposure to cadmium: A major public health concern. Geneva: WHO, 2010. Available at: <http://www.who.int/ipcs/features/cadmium.pdf>, last access: September 2015.
3. Brenna, J. T., Varamini, B., Jensen, R. G., Diersen-Schade, D. A., Boettcher, J. A. and Arterburn, L. M. (2007). Docosahexaenoic and arachidonic acid concentrations in human breast milk worldwide. *American Journal of Clinical Nutrition*, 85, 1457-1464
4. Brace, W. H., Carol, L. W., Cynthia, R. H., Myla, E., Judy, R. S., *et al.* (2015). Maternal versus infant vitamin D supplementation during lactation: a randomized controlled trial. *Pediatrics*, 136(4), 625-634
5. Signe, B., Lotte, N. J., Xiaole, Z., Steffen, H., Hiroshi, M. U., *et al.* (2018). Osteopontin levels in human milk vary across countries and within lactation period: data from a multicenter study. *Journal of Pediatrics*, 67(2), 250-256
6. Fyनेface, C. A., Joel, B. B. K and Felix, E. K. (2020). Assessment of creatinine levels in blood and saliva of haemodialysed subjects. *International Journal of Advances in Nephrology and Research*, 3(1), 21-25
7. Fyनेface, C. A., onengiyeofori, I. and Davies, T. E. (2018). Evaluation of saliva for monitoring renal function in haemodialysed patients at university of port Harcourt teaching hospital. *Asian Journal of Biochemistry, Genetics and Molecular Biology*, 1(2), 1-6
8. Mangel, L., Ovental, A., Batscha, N., Arnon, M., Yarkoni, I. and Dollberg, S. (2015). High Fat Content in BreastmilkExpressed Manually: A Randomized Trial. *Breastfeed Med.*, 10(7), 352-354
9. Andreas, N. J., Kampmann, B., Mehring. and Le-Doare, K. (2015). Human breast milk: A review on its composition and bioactivity. *Early Human Development*. 91(11), 629–635.
10. Matylda, C. L., Barbara, K. O. and Magdalena, O. P. (2018). Breast Milk Macronutrient Components in Prolong Lactation. *Nutrients*, 10(12), 1893