**ABSTRACT**

Amino acids are the building blocks of protein and as such are important in breast milk for infant’s growth and development. This study was aimed at evaluating the protein and amino acid composition of breast milk of lactating mothers in Umuahia, Abia State. Breast milk samples were collected from 27 lactating mothers during 2-4 weeks of lactation. Dietary intake was determined using weighed inventory method and 24 hour dietary recall. Proximate analysis was carried out using standard methods. Amino acid was by the use of amino acid analyser. Data was analysed using means (SD) and t-test. Results showed that mothers had a mean age of 26.0±0.44yrs, weighed 72.60±11.20kg and had height of 1.61±0.08cm. Protein, fat, carbohydrate and energy intake were 57.08±11.86g, 45.76±31.65g, 459.88±104.59g and 2479.64±223.40kcal, respectively. Cereals and legumes were the main sources of protein. Protein content of breast milk was 1.09±0.38g/100ml. The main amino acids in the breast milk of the mothers were leucine and aspartic acid, while cysteine was the least. Correlation analysis between protein in diet and that in breast milk showed no significant association (P>0.05). Amino acid levels were similar to that reported from other developing countries.

**Keywords:** Amino acids, breast milk, infants, protein, composition.

**INTRODUCTION**

Amino acids are the building blocks of protein, as such, they are important in breast milk for infant growth and development. Amino acids perform other functions, such as facilitating digestion by increasing uptake of other nutrients and enhancing the infant’s immune function against pathogenic bacteria, viruses and yeasts [1]. The protein content of breast milk has been shown to range from 1.4-1.6g/100ml during early lactation, 0.8-1.0g/100ml by 3-4 months of lactation and 0.7-0.8g/100ml after 6 months of lactation [2 and 3].

Studies on the amino acid composition of breast milk have been carried out in different parts of the world. However, very few studies have been carried out in Nigeria [4]. This study will therefore examine the amino acid composition of breast milk with a view of finding the abundant and limiting amino acids in breast milk of mothers in Umuahia, Nigeria.
MATERIALS AND METHODS

A total of 27 lactating mothers within 2-4 weeks postpartum were randomly selected from a group of mothers attending immunization clinics at the Federal Medical Centre, Umuahia. Inclusion criteria included mothers who had healthy term babies, while mothers who were ill or had ill babies were excluded from the study. Both oral and written consent were obtained from the mothers before commencement of the study. Ethical clearance was obtained from the ethics committee of the Federal Medical Centre, Umuahia.

Collection of breast milk samples
Breast milk samples (5ml) were collected between 2-4 weeks of lactation. This was expressed with a manual pump into sterile containers provided by the investigator. They were placed on ice and transported to the laboratory. Samples of breast milk collected were pooled and frozen at 4°C for analysis.

Dietary intake assessment
Dietary intake assessment involved a three day weighed inventory analysis (2 week days and a weekend day) and 24 hour recall of foods consumed as described by Ukegbu [5]. Food composition tables [6] were used to calculate the energy and macronutrient of common foods and snacks. Duplicate samples of traditional foods and meals which could not be found in Food Composition Tables [6] were collected from the homes of the subjects, placed in plastic bags and transported in insulated containers with ice to the laboratory.

Proximate analysis
Protein was analysed using the Kjeldahl method, fat was by Soxhlet extraction method [8], carbohydrate was calculated by difference, while energy was calculated using Atwater factors of 4, 4 and 9 kcal for protein, carbohydrate and fat, respectively [8].

Amino acid analysis
Amino acid analysis of breast milk samples was carried out using methods described by Speckman et al. [7]. A known sample of breast milk was dried to constant weight, defatted, hydrolysed, evaporated in a rotary evaporator and loaded into the Technicon Sequential Multi-sample Amino Acid Analyser (TSM). This analysis was carried out at the Zoology Department of the University of Jos, Nigeria.

Statistical analysis
Data was analysed using SPSS version 15. Values were expressed as means (SD), percentages and frequencies. Pearson’s correlation coefficient was performed to determine the relationship between protein intake and its concentration in breast milk. P<0.05 was regarded as statistically significant.

RESULTS AND DISCUSSION

The mean age of the mothers was 26.0±0.44 years. Their height, weight and BMI were 1.61±0.08 cm, 72.60±11.20 kg and 28.11±4.41 kg/m², respectively.

The results of the energy and protein intake of the mothers are presented in Table 2. The results revealed that the energy intake of the mothers was 2479.64±223.40 kcal. Protein was 57.08±11.86 g, fat was 45.76±31.65 g and carbohydrate 459.88±104.59 g. Protein accounted for about 9% of total energy intake, while fat and carbohydrate provided about 17% and 73% of total energy intake. The energy and protein intake of the mothers were insufficient, while the mothers had adequate fat and carbohydrate intake. The food sources of protein from the dietary intake assessment of the mothers were mainly from grains, roots and tubers, vegetables and fruits. These are poor sources of essential amino acids (EAA) and do not have a similar amino acid pattern when compared to animal protein. The mothers ate little pieces of meat, fish, eggs and soy which are good sources of essential amino acids. The insufficient protein intake could thus be attributed to the low protein content of their diets. The protein sources were mainly from cereals and legumes which have low lysine and cysteine content, respectively. This may have probably resulted in low lysine and cysteine content of their breast milk. Again, inadequate consumption of animal food sources could have further resulted in low essential amino acid content of breast milk of the mothers since these were plant based.
Mean energy value of breast milk was 69±0.01 kcal/100 ml, while protein was 1.09±0.38 g/100 ml. Fat and carbohydrate were 3.32±0.17 g/100 ml and 6.75±0.05 g/100 ml. The mean protein content of breast milk was in line with standards (0.8-1.2 g/100 ml) for mature milk [9]. Similarly, fat, carbohydrate and energy were in line with standard range [9].

The correlation between protein in diet and that in breast milk was not significantly different (p<0.05) (Table 4). This indicates that protein content in the diet did not have any direct relationship with that in breast milk. This supports findings of similar studies [10,11].

Table 5 summarizes the amino acid composition of breast milk of the mothers involved in the study. The total amino acid was 72.4 g/100 g. Leucine (10.02 g/100 g) and aspartic acid (9.85 g/100 g) were the major amino acids, while cysteine (0.99 g/100 g) was the lowest. It was observed that concentration of most of the amino acids were somewhat higher than standard, except for lysine, threonine, cysteine, valine and proline which were lower [12]. The values obtained in this study for all the amino acids were higher than those reported by Nnayelugo et al. [4], except for phenylalanine which was higher in their own study (5.88 g/100 g). It is possible that the protein content of diets of mothers involved in this study were of higher quality than theirs. It could also be that the methods of analysis have improved over the years. Similarly, a study [13] reported that poorly nourished mothers in Pakistan secreted milk low in methionine and cysteine and attributed it to poor protein quality in the diet of the mothers they studied. The limiting amino acids (lysine, threonine and valine) may be explained by low consumption of animal protein and soy which are good sources of the essential amino acid lysine. Erdman and Fordyce [14] and Xiao [15] reported that soy is inexpensive and contains adequate quantities of essential amino acids. Inadequate protein intake of the mothers could have also contributed to the limiting amino acids observed. Although, the amino acid concentration of the women’s breast milk was adequate quantitatively, it was not qualitatively similar to the amino acid pattern of animal proteins. Therefore, the protein quality of the mothers’ diet needs to be improved upon.
Table 5: Amino acid composition (g/100g) of breast milk of mothers involved in the study

<table>
<thead>
<tr>
<th>Amino acids</th>
<th>Amount (g/100g)</th>
<th>FAO/WHO/UNU (1985) Standards (g/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lysine</td>
<td>6.33</td>
<td>11.1</td>
</tr>
<tr>
<td>Threonine</td>
<td>4.22</td>
<td>8.6</td>
</tr>
<tr>
<td>Valine</td>
<td>5.17</td>
<td>9.4</td>
</tr>
<tr>
<td>Methionine</td>
<td>2.94</td>
<td>1.3</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>6.09</td>
<td>4.0</td>
</tr>
<tr>
<td>Leucine</td>
<td>10.02</td>
<td>8.9</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>4.48</td>
<td>3.2</td>
</tr>
<tr>
<td>Histidine</td>
<td>2.50</td>
<td>2.6</td>
</tr>
<tr>
<td>Cysteine</td>
<td>0.99</td>
<td>1.2</td>
</tr>
<tr>
<td>Tyrosine</td>
<td>4.19</td>
<td>2.6</td>
</tr>
<tr>
<td>Arginine</td>
<td>3.91</td>
<td>3.9</td>
</tr>
<tr>
<td>Aspartic acid</td>
<td>9.85</td>
<td>6.8</td>
</tr>
<tr>
<td>Serine</td>
<td>3.60</td>
<td>3.2</td>
</tr>
<tr>
<td>Alanine</td>
<td>5.03</td>
<td>4.2</td>
</tr>
<tr>
<td>Proline</td>
<td>3.08</td>
<td>10.2</td>
</tr>
</tbody>
</table>

REFERENCES