

BIOCHEMICAL STUDIES ON HUMAN MILK FROM ABEOKUTA, NIGERIA

M. KADIRI, and O. O. OLEMBE

(Received 24 October 1997; Revision accepted 27 February 1998)

ABSTRACT

Biochemical analysis was carried out on the human milk of lactating mothers, aged 15-40 years from Abeokuta, Nigeria. Microbiological sampling showed the human milk to be microbiologically pure for infant consumption as no viable bacteria growth per ml was found to be nil. The samples had pH values ranging from 5.8 to 6.2, protein content ranged from 14.3 to 21.4mg/ml, calcium content ranged from 0.13 to 0.27mg/ml and sugar content ranged from 1.6 to 3.0mg/ml. Women aged, 15-20 years had the most nutritive human milk profile, based on these parameters ($P < 0.05$).

KEY WORDS: Biochemical Studies, human milk, Nigeria.

INTRODUCTION

Milk is the natural food of the newborn mammal whom it is the sole source of food and nourishment some period after birth. Milk is the secretion of the mammary glands of animals which suckle their young (Okoronye and Ngoddy, 1985). An increasing number of women are currently choosing to breast feed their infants for longer periods (Martinez and Nalesiensi, 1981).

Milk is a good source of protein of high biological value as well as calcium. It therefore supports sound bone and teeth development as well as growth in children. Useful vitamins: riboflavin, thiamin, biotin, niacin, pantothenic acid and vitamin B12 have been reported in human milk (Casey and Nisbidge, 1983).

Milk is known to protect infants against bacterial infection and allergic reactions (Ebrahim, 1983; Ogunson, 1994). Water-soluble vitamins, fatty acid composition and lipid contents of human milk have been shown to reflect maternal food intake (Soderhjelm, 1953; Insull and Ahrens, 1959; Belavady, 1978; Ogunson et al, 1992; Ogunson, 1994). As an example, Borschel et al (1986) in his comparative study of the fatty acid composition of mature human milk of Nigerian and American women found that breast milk of Nigerian women contained higher percentages of capric, myristic, linoleic and arachidonic acids while the reverse of higher percentages of stearic and oleic acids in breast milk of American women was true. In the same vein, breast milk of American and Australian women were found to contain linoleic acid values of 14% and 11% respectively (Janssohn et al, 1981; Ogunson and Kneebone, 1981; Bitaman et al, 1983).

The high nutritive value of milk makes it rapidly contaminated by microorganisms and thus gives it a short shelf life. The present study was undertaken with the following aims in view: to assess the nutritional value of human milk from Abeokuta, Nigeria; to ascertain the purity of the milk and to compare the nutritive values of human milk produced by lactating mothers of varying ages from this region.

MATERIALS AND METHODS

Human milk collection: Human milk samples were collected from 250 healthy lactating mothers, aged 15 to 40 years by manual expression, from 5 randomly selected hospitals in Abeokuta, the capital of Ogun

State of Nigeria. The lactating mothers were within one month of having given birth. Twenty millilitres of human milk were obtained from each lactating mother and delivered into sterile sampling bottles which were immediately frozen prior to analysis. The lactating mother's nipple was thoroughly cleaned with 70% alcohol prior to milk collection.

Microbiological Analysis of Human Milk: Serial dilutions from 10^{-1} to 10^{-3} of each 1ml milk sample were prepared. For the viable bacterial count, 0.1ml of the stock dilutions, 10^{-1} to 10^{-3} were plated in triplicate on nutrient agar plates. The inoculum was spread evenly round the plate using pour plate method. The plates were incubated at 37°C for 48h after which bacterial colony presence in the plates was ascertained.

Calcium Content and pH Value of Human Milk: Calcium content was determined by pipetting 1ml of the milk sample into a beaker, diluting it to 100ml, adding 2 drops methyl red and ammonium hydroxide added drop-

wise until a brownish orange colour was obtained. Thereafter, 2 drops of dilute hydrochloric acid were added and the solution diluted with 50ml of water. The solution was boiled and 10ml of hot 4.2% ammonium oxalate solution added with stirring until a precipitate was formed. The precipitate was filtered out, washed with 40% ammonium hydroxide solution and dissolved in a mixture of 125ml water and 5ml 98% sulphuric acid. The ensuing solution was heated to 70°C using a water-bath and titrated against 0.05N potassium permanganate solution (IITA, 1979). The pH of the milk samples was determined by dipping the electrode of a warmed pH meter in 10ml human milk sample and the pH noted.

Protein and Sugar Contents of Human Milk: Protein content was determined using Folin Ciocalteu method by adding 3ml of solution A to 1ml of human milk (solution A consisted of 50ml 2% Na₂CO₃ in 0.1M NaOH mixed with 1ml of a mixture of 10% CuSO₄·5H₂O and 2% sodium tartarate). At 10 min. after addition, 1ml of Folin Ciocalteu reagent was added and 30min. later, the optical density of the resulting solution was taken at 570nm against a blank that had 1ml human milk replaced with 1ml distilled water. The protein standard curve was prepared from various concentrations of casein in 0.1M NaOH (Lowry et al, 1951). Sugar content was determined using phenol-sulphuric acid method of Dubois et al (1956) by adding 1ml of 5% phenol and 5ml concentrated H₂SO₄ to 1ml of human milk and the optical density taken against a blank at 490nm. The blank contained 1ml of distilled water instead of milk extract and the standard curve was prepared using various concentrations of glucose (Dubois et al, 1956).

RESULTS AND DISCUSSION

The pH values of the human milk ranged from 5.9 to 6.3 while the protein contents ranged from 14.3 to 21.4mg/ml (Table 1). With regards to calcium and sugar contents, the least values of 0.13mg/ml and 1.6mg/ml were observed in age group 36-40 while age

group 15-20 had the highest values of 0.27mg/ml and 3.0mg/ml (Table 1)

Bacterial growth in the cultured human milk was found to be nil. This result is in agreement with the report of American Academy of Pediatrics which considered human milk to be a clean and optimal source of nutrition for the infants (American Academy of Pediatrics, 1978). Protein, calcium and sugar contents of human milk were observed to decline in quantities with increase in age of the women while the converse was true with regards to pH values (Table 1). Calcium and sugar contents of 15-20 years age group's breast milk were significantly higher than those of other age groups ($P < 0.05$) whereas in the case of protein contents, only those of age groups 31-35 and 36-40 were significantly lower to that of age group 15-20 years (Table 1). The implication of the above findings is that women, aged 15-20 years produce the most qualitative human milk.

REFERENCES

- American Academy of Pediatrics. 1978. Committee on nutrition commentary on breast-feeding and infant formulas, including proposed standard for formulae. *Pediatrics* 67: 278-285.
- Belavady, B. 1978. Lipid and trace element composition of human milk. *Acta Paediatr. Scand.* 67: 566-571.
- Bitaman, J., Wood, D.L., Hamosh, M., Hamosh, P. and Mehta, N.R. 1983. Comparison of the lipid composition of breast milk from mothers of term and pre-term infants. *Amer. J. Clin. Nutr.* 38: 300-312.
- Borschell, M.M., Elkin, R.G., Kirksey, A., Sory, J.A., Galal, O., Harrison, G.G. and Jerome, N.W. 1986. Fatty acid composition of Egyptian and American women. *Amer. J. Clin. Nutr.* 44: 330-335.
- Casey, C.E. and Hambidge, C.F. 1983. Nutritional aspects of human lactation. *Amer. J. Clin. Nutr.* 41: 1193-1197
- Dubois, M., Giles, A.K., Hamilton, K.J., Rebers, A.P. and Smith, F. 1956. Colorimetric method for

Table 1: Mean pH, protein, calcium and sugar contents of human milk from lactating mothers, aged 15-40 years.

Age group (years)	pH Level	Protein content (mg/ml)	Calcium content (mg/ml)	Sugar content (mg/ml)
15 - 20	5.9	21.4	0.27	3.0
21 - 25	6.1	19.8	0.20	2.6
26 - 30	6.1	18.5	0.18	2.4
31 - 35	6.2	17.5	0.16	2.0
36 - 40	6.2	14.3	0.13	1.6

- determination of sugar related substances. *Anal. Chem.* 28: 350-354.
- Shim, C.J. 1983. Nutrition in mother and child health. *Macmillan tropical community health manual*, New York 14: 14-15.
- Umarso, J., Cockburn, F. and Patrick, W.A. 1992. Infant cerebral cortex phospholipids fatty acid composition and diet. *The Lancet* 340: 180.
- Wason, R.A. and Kneebone, G.M. 1981. Fatty acid composition of human colostrum and mature breast milk. *Amer. J. Clin. Nutr.* 34: 252-257.
- Oronje, A.L. and Ngoddy, P.O. 1985. *Integrated food science and technology for the Tropics*, Longmans (London), pp. 350-367.
- W.A. 1979. Selected methods for soil and plant analysis. *Inter. Inst. of Tropic. Agricul. Manual Series No.1*, IITA, Ibadan, Nigeria, pp. 47-51.
- Hill, W.Jr. and Ahrens, E.H. Jr. 1959. The fatty acids of human milk from mothers on diet taken ad libitum. *Biochem. J.* 72: 27-33.
- Jansson, L., Akesson, B. and Holmberg, L. 1981. Vitamin E and fatty acid composition of human milk. *Amer. J. Clin. Nutr.* 34: 8-13.
- Lowry, O.H., Rosebrough, H.N., Farr, L.A. and Randall, J.R. 1951. Protein measurement with the Folinphenol reagent. *J. Biol. Chem.* 193: 263-275.
- Martinez, G. and Nalezienski, J. 1979. The recent trend in breast feeding. *Pediatrics* 64: 686-692.
- Martinez, G. and Nalezienski, J. 1981. 1980 update: the recent trend in breast feeding. *Pediatrics* 67: 260-263.
- Ogunsanya, O.M. 1994. Level of zinc in human milk of selected women in Abeokuta. Final diploma dissertation of University of Agriculture, Abeokuta, pp. 59-60.
- Soderhjelm, L. 1953. Fat absorption studies. VII Polyunsaturated fatty acids in human milk and their variation with dietary fat. *Acta Soc. Med. Upsal.* 58: 244-251.