

ISOTOPIC EVALUATION OF BREAST MILK INTAKE, ENERGY METABOLISM, GROWTH AND BODY COMPOSITION OF EXCLUSIVELY BREAST FED INFANTS IN PAKISTAN

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Abstract

There is considerable evidence to support the view that the current growth standards for infants, which are in-use globally, may be inappropriate. This is based on the observation that these were derived from largely formula-fed western populations and recent studies documenting that exclusively breastfed young infants exhibit a lower growth trajectory. However, there are few studies objectively evaluating energy metabolism, body composition and growth in exclusively breastfed infants, and none in developing countries. We propose to evaluate this longitudinally in an appropriate sample of exclusively breast fed newborn infants in Pakistan. These newborn infants will be well characterized at birth and sequential measurements of breast milk intake and energy expenditure will be made using doubly labeled water, big-impedance analysis and indirect calorimetry.

1. SCIENTIFIC BACKGROUND AND SCOPE OF THE PROJECT

The NCHS-WHO growth reference standards [1] are widely used as reference growth centiles in the developing world. Despite numerous efforts to define separate regional or national growth standards which tend to accept lower birth weight and growth trajectories as normal [2,3], recent evidence indicates that it may be inappropriate to do so [4]. However, the following major concerns have been expressed about the current/NCHS WHO reference standards.

The standards were drawn from a single, ethnically homogenous Caucasian population of artificially fed infants.

Measurements were taken at wide intervals with outdated curve-fitting procedures.

Several studies of exclusively breastfed infants have revealed lower growth trends in comparison with NCHS/WHO reference standards.

A major multicentre international growth reference study is presently underway to study the growth of exclusively or predominantly breastfed children longitudinally for the first 24 months of life. This standard will be drawn from a comparable multiethnic population of children in different parts of the world and may provide a valid international reference standard. This infant growth data from 6 to 8 sites in different countries will be combined with cross-sectional data from children aged 18-71 months, in order to construct a new reference growth standard.

While several studies have established the immunological advantages of exclusive breastfeeding, reduced infectious disease morbidity [5,6] and improved neurological outcome [7], few have addressed the issue of body composition. In drawing up these new standards, it will be important to document that these breastfed infants, even though following a lower growth trajectory [8], are normal. Similarly it will be important to correlate growth and variations thereof, to the absolute breast-milk intake of the reference population. Classically breast milk intake has been estimated by test weighing [9] but this is a cumbersome technique and impractical for such evaluation in the community. The recently developed methods of evaluating breast milk intake by using stable isotopes like deuterium ($^2\text{H}_2\text{O}$) [10-12] and measurement of energy metabolism by using the doubly labeled water ($^2\text{H}_2^{18}\text{O}$) [13-16], offer the exciting possibility of non-invasive evaluation of the relationship of human lactation and growth.

2. METHODS

A consecutive 150 mothers-infant pairs will be prospectively identified for inclusion in the study on the basis of fulfillment of inclusion criteria, motivation to exclusively breastfeed for 6 months and ease of monitoring. It is anticipated that of these at least 100 will fulfill criteria of exclusively breastfeeding for 6 months and appropriate introduction of complementary foods thereafter.

We shall randomly select 10 mothers from the above 100 mothers-infant dyads for detailed evaluation of lactation performance and body composition/energy metabolism by using doubly labeled

water ($^2\text{H}_2^{18}\text{O}$) as per the following protocol. The anthropometric measurements will be carried out at fortnightly and monthly intervals as per the International Growth Reference Study Protocol. Actual breast milk intake and energy expenditure will be estimated in a randomly selected subset of exclusively or predominantly breastfed infants by administration of doubly labeled water at time points A, B, C (Figure 1).

Days	15-30	90-105	165-180
Administration of $^2\text{H}_2^{18}\text{O}$ to mother, baby & collection of salivary samples	↑	↑	↑
	A	B	C

In addition, we shall measure body composition and energy metabolism at these time points by multi-frequency bio-impedance analysis (using the Xitron Hydra®) and indirect calorimetry using the Deltatrac II® metabolic monitor. The isotopic concentrations in salivary samples will be measured by isotope ratio mass spectroscopy (IRMS). In addition, we shall measure ^2H concentration by indirect spectrophotometry using the Miran I FF indirect spectrophotometer, and validate it against IRMS. Preliminary data from field studies in Indonesia suggest that the method is valid (Caballero B. persona communication, 1998).

3. DETAILED RESEARCH OBJECTIVES

We would aim to study:

3.1. The relationship of breast milk intake and its micronutrient composition with growth in early infancy

There is considerable interest in the relationship of micronutrient status and growth in early infancy, especially in the link of zinc and growth faltering [17, 18]. Preliminary data from Pakistan also indicates a possible link of zinc deficiency with low birth weight [19] (Figure 2). The relationship of micronutrient composition and growth will be evaluated by estimating zinc and iron content of breast milk (by atomic absorption spectrophotometry) on samples obtained at corresponding time points.

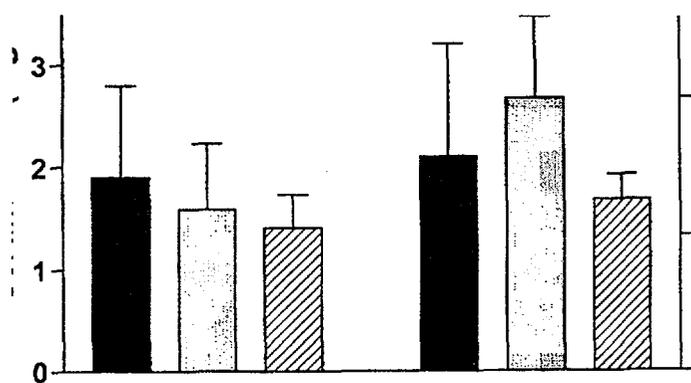


Figure 2: Serum retinal binding protein (RBP) and plasma zinc & copper in Pakistani infants according to relative size at birth.

There is considerable interest in the relationship of micronutrient content and bioavailability from breast milk and growth in early infancy. While the coexistence of maternal micronutrient especially zinc deficiency and intrauterine growth retardation in developing countries is well recognized, its contribution to postnatal growth restriction is less well established. In an important study in urban Paris, Walravens et al [20] demonstrated improved growth in normal infants after zinc supplementation, although their findings were questioned [21]. Much of the zinc in maternal breast milk is highly bioavailable and the relationship of breast milk zinc content and its absolute intake with postnatal growth is of considerable interest.

3.2. To evaluate the relationship of energy metabolism and growth in early infancy

There has been considerable interest in immuno-stimulation and growth faltering in developing countries, but few longitudinal studies have attempted to scientifically evaluate this possibility. Data from the recently concluded CRP on the use of stable isotopes to assess amino acid metabolism following immuno-stimulation strongly suggest this possibility. It will be extremely useful to correlate dynamic studies of energy metabolism, coexisting morbidities and growth during this longitudinal study.

There are few studies correlating growth during early infancy with energy metabolism in developing countries. While studies employing indirect calorimetry are clearly impractical for field studies, the doubly labeled water method is uniquely suitable for field-based studies. Of an estimated 250 studies evaluating this technique globally, only about 12 have been performed in developing country situations and of these only 4 have addressed issues of infant nutrition [22].

3.3. To evaluate the body composition of reference study infants by skin fold thickness measurements and bio-impedance analysis

While exclusively breastfed infants are recognized to be leaner than formula fed infants, it will be important to demonstrate "normal" body composition in the infants determining the new WHO International reference standards. Our studies employing bio-impedance analysis, as well as skin fold thickness measurements, will aim to obtain this data in a standardized fashion and correlate it with estimates from the doubly labeled water method.

Although traditionally body composition studies using anthropometry or bio-impedance analysis have been used in ambulatory patients, there are concerns about their validity in field situations. This study will attempt to correlate these two types of measurements and will be the first study in Pakistan.

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