

ESTABLISHMENT AND APPLICATION OF DEUTERIUM DILUTION METHOD FOR MEASURING BREAST MILK INTAKE OF PAKISTANI INFANTS

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Abstract

*This project will be a collaborative study between the Pakistan Institute for Nuclear Science and Technology (PINSTECH) and Department of Pediatrics, Pakistan Institute of Medical Sciences (PIMS). It may also involve the Polyclinic, Islamabad. Pediatricians who have had experience in lactation management clinic and hence breast-milk intake studies by conventional methods (test weighing) will be formally involved in the study. Human milk intake and growth performance of exclusively breast-fed infants will be monitored over the first six months of life. The deuterium dilution method will be used for measuring breast-milk intake and will be compared with the test weighing method. The growth of exclusively breast-fed children will be compared with the NCH growth standards. Records of illnesses that may lead to sub-optimal growth will be kept. Episodes of diarrhoea, in particular, will be correlated with growth faltering. Acquisition of *Helicobacter Pylori* detected by the use of the ^{13}C Urea breath test at three month intervals during the study period examined for relationships with diarrhoea.*

*Thus, this study will not only provide preliminary data on breast-milk intake (based on a more accurate isotopic method as compared to test weighing) but also on the growth patterns of exclusively breast-fed infants. Further, information on the significance of *Helicobacter Pylori* to infection with episodes of diarrhoeal infection and growth faltering.*

1. SCIENTIFIC BACKGROUND AND SCOPE OF THE PROJECT

While PINSTECH has good analytical facilities in IRMS they have to be developed for clinical and physiological studies. In particular, sample preparation systems, specifically for use in this work, and separate from those used in isotope hydrology are required. When this has been done the laboratory will be in a position to apply stable isotope techniques in areas relevant to this CRP, namely the relationship of breast-milk intake to growth in Pakistani infants and the significance of diarrhoeal disease to growth faltering and the putative links with the acquisition of *Helicobacter Pylori* infection¹.

2. METHODS

2.1. Subjects

The identification and selection of suitable subjects will be made by the trained regular staff of MCH centre, affiliated with Polyclinic Hospital, Islamabad. The enrolment will be made during antenatal visits. The investigators will then explain the purpose of the study and ethical issues to mothers willing to breast-feed. All mothers with positive consent will be

¹ WEAVER, L.T., Aspects of *Helicobacter pylori* infection in the developing and developed world. *Helicobacter pylori* infection, nutrition and growth of West African infants, *Trans R Soc Trop Med Hyg*, **89** (1995) 347-350.
DALE, A., THOMAS, J.E., DARBOE, M.K., COWARD, W.A., HARDING, M., WEAVER, L.T., *Helicobacter pylori* infection, gastric acid secretion, and infant growth. *J Pediatr Gastroenterol Nutr* **26** (1998) 393-397.

recruited for the study and will be followed up at their homes. The selection criteria will be that the mothers:

- should be healthy
- live in the vicinity of Islamabad/Rawalindi area (and are thus within reach of the project team)
- are willing to be visited
- are willing to continue breast feeding.
- be in their second or later pregnancy
- had no difficulties at labour

The babies will be normal with birth weight > 2.5 kg. Approximately 20-25 subjects will be recruited.

2.2. Equipment and field work

The MCH centre is a 60 bed maternity unit and has all the facilities necessary for anthropometric measurements on mothers and babies. Field workers trained to collect milk samples from the mother and saliva or urine from the babies will be recruited. The field workers will also make the anthropometric and other measurements required for the study (age, weight, height, skin fold thickness, dietary and morbidity records). Breast-milk intake measurements will be made on alternate months up to one year using the "dose to the mother method"² Comparisons will be made between the isotope method and a test-weighing procedure. In addition, the urea breath test for *Helicobacter Pylori* will be administered.

2.3. Mass spectrometric measurements

All IRMS measurements will be made at PINSTECH. While the procedures for ¹³C analyses in breath samples are routine, preparation line facilities for hydrogen preparation from water samples are only available for hydrology and additional lines have to be built. IRMS measurement of D/H ratios in gas samples is subsequently routine.

2.4. Results

The outcomes of the study are expected to be available in 2000, with most of the studies being performed in 1999. In 1998 the mass-spectrometric methods will be established.

² BUTTE, N. F., WONG, W. W., PATTERSON, B. W., GARZA, C., KLEIN, P. D., Human-milk intake measured by administration of deuterium oxide to the mother: A comparison with the test-weighing technique, *Am. J. Clin. Nutr.* **47** (1988) 815-821.

ORRWING, A. K., HEYWOOD, P. F., COWARD, W. A., Longitudinal measurements of breast milk output by a ²H₂O tracer technique in rural Papua New Guinean women, *Hum. Nutr. Clin. Nutr.* **40** (1986) 451-467.