

Effects of prenatal food and micronutrient supplementation on infant development: a randomized trial from the Maternal and Infant Nutrition Interventions, Matlab (MINIMat) study^{1–3}

Fahmida Tofail, Lars Åke Persson, Shams El Arifeen, Jena D Hamadani, Ferdina Mehrin, Deborah Ridout, Eva-Charlotte Ekström, Syed N Huda, and Sally M Grantham-McGregor

ABSTRACT

Background: Few data exist for the effects of multiple micronutrient (MM) or food supplementation to undernourished pregnant women on their offsprings' development.

Objective: We aimed to compare the effects on infant development of early (8–10 wk gestation) or usual (\approx 17 wk gestation) supplementation with food and MM, 30 mg Fe + 400 μ g folate, or 60 mg Fe + 400 μ g folate.

Design: A large, randomized, controlled trial of pregnancy supplementation was conducted in Bangladesh. A subsample of infants ($n = 2853$) were assessed on 2 problem-solving tests (support and cover tests), the motor index of the Bayley Scales of Infant Development, and Wolke's behavior ratings at 7 mo of age.

Results: There were no significant effects of any intervention in the group as a whole. However, infants of undernourished mothers [body mass index (BMI; in kg/m^2) < 18.5] who received early food supplementation performed slightly but significantly ($P = 0.035$) better on the support test than did infants of mothers who received usual food supplementation (z score: 0.17; 95% CI: 0.01, 0.33). There were no benefits in infants of higher-BMI mothers ($P = 0.024$ for BMI \times food interaction). Children of low-BMI mothers who received MMs had slightly better motor scores (z score: 0.28; 95% CI: 0.08, 0.48) and activity ratings (z score: 0.24; 95% CI: 0.037, 0.45) than did those who received 30 mg Fe + 400 μ g folate, whereas other children did not benefit ($P = 0.05$ for both motor scores and BMI \times micronutrients and for activity and BMI \times micronutrients).

Conclusions: Small benefits from early food and MM supplementation were found in infants of low-BMI but not of high-BMI mothers. However, the benefits were of doubtful functional importance, and longer follow-up is required to determine programmatic implications. *Am J Clin Nutr* 2008;87:704–11.

KEY WORDS Maternal nutrition, pregnancy, food and micronutrient supplementation, problem-solving test, psychomotor development, Bangladesh, infants, body mass index

INTRODUCTION

The prevalence of low birth weight [(LBW) ie, <2.5 kg] is high in developing countries; it is estimated to be 36% in Bangladesh (1). Maternal undernutrition is an important cause of LBW, and 38% of Bangladeshi women are undernourished (1).

Deficiencies in several micronutrients are also common in pregnancy (2), and it has been suggested (3, 4) that supplementing pregnant women with multiple micronutrients (MMs) may have greater benefits in their offspring at 1–6 y old than does the current World Health Organization recommendation of using iron folate (5). A recent Cochrane review of 13 trials found that balanced protein-energy supplementation during pregnancy moderately increased the birth weight of the offspring (6), whereas other investigations showed the effect of multiple micronutrients to be inconsistent (7–11).

There is concern that deficiencies of macronutrients (12–14) and micronutrients (15–17) during pregnancy also may detrimentally affect the neurodevelopment of offspring. Animal research has shown that protein-energy deficiency during gestation may cause permanent alteration of brain structures and impaired cognitive function (18). In humans in developed countries, food supplementation in apparently healthy pregnant women had no effect on children's development (19–21), whereas studies from developing countries showed mixed results. In Taiwan, supplementation of marginally undernourished women throughout

¹ From the International Centre for Diarrhoeal Disease Research, Bangladesh, Dhaka, Bangladesh (FT, SEA, JDH, and FM); International Maternal and Child Health, Uppsala University, Uppsala, Sweden (LAP and E-CE); the Institute of Nutrition and Food Science, University of Dhaka, Dhaka, Bangladesh (SNH); and the Centres for International Child Health and the Centre for Paediatric Epidemiology and Biostatistics (DR), Institute of Child Health, University College London, London, United Kingdom (SMG-M).

² Supported by The International Centre for Diarrheal Disease Research, Bangladesh; the UK Medical Research Council; the Swedish Research Council; the UK Department for International Development; the Global Health Research Fund, Japan; the Child Health and Nutrition Research Initiative; Uppsala University; the US Agency for International Development, under the Cooperative Agreement #388-G-00-02-00125-00; the Australian International Development Agency; the Government of Bangladesh; the Canadian International Development Agency; The Kingdom of Saudi Arabia; the Government of the Netherlands; the Government of Sri Lanka; the Swedish International Development Cooperative Agency; and the Swiss Agency for Development and Cooperation.

³ Reprints not available. Address correspondence to F Tofail, Child Development Unit, Clinical Sciences Division, ICDDR,B, 68 Shaheed Tajuddin Ahmed Sharani Mohakhali, Dhaka 1212, Bangladesh. E-mail: ftofail@icddr.org.

Received June 4, 2007.

Accepted for publication October 1, 2007.

pregnancy and lactation benefited their infants' motor but not mental development (22) or later intelligence (23). In Colombia, supplementation during the last trimester of pregnancy did not benefit the children's mental development (24).

Information on the effect of MM deficiencies in pregnancy on children's development is limited (25). Supplementing women with either 16 or 4 micronutrients during the periconceptional period made no difference to the offsprings' cognitive development (26); however, the children whose mothers had received 16 micronutrients had lower anxiety at age 6 y (15).

A multicenter trial of the effect of giving 15 MMs during pregnancy on birth outcomes was launched in 6 countries (4). The trial in Bangladesh was integrated into the Bangladesh Integrated Nutrition Program (BINP), which provided food supplementation during pregnancy through community nutrition centers. This trial aimed to compare the effect on birth outcomes of MM supplementation during pregnancy with that of different doses of iron and folate supplementation (30 or 60 mg Fe in addition to 400 μ g folate) and the effect of early (ie, periconceptual) food supplementation with that of usual food supplementation during pregnancy.

We took the opportunity to examine the effects of the supplements on children's cognitive and motor development and behavior at age 7 mo in a subsample. We hypothesized that the above treatment would benefit the infants of malnourished mothers more than those of better-nourished mothers.

SUBJECTS AND METHODS

Location

The study was conducted in Matlab, a poor rural subdistrict in the east central plain of Bangladesh. The main economic activities of the area are farming and fishing, and \approx 85% of the population is Muslim. The International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B) has a Health and Demographic Surveillance System that has recorded vital demographic information in the area since 1966.

Intervention

With the use of the Health and Demographic Surveillance System, all pregnant women were identified within 6–8 wk of conception through regular home visits, and their pregnancies were confirmed by urine tests and ultrasonography. After enrollment, women were randomly assigned to 6 intervention groups—2 food groups, each of which was subdivided into 3 micronutrient groups.

Food

In the early-food group, women were urged to take supplementation as soon as pregnancy was identified, whereas, in the usual-food group, women were left to choose when to start. Food packets containing roasted rice powder (80 g), roasted lentil powder (40 g), molasses (20 g), and soy oil (12 mL) provided 600 kcal (2512 kJ) of energy. All enrolled pregnant women were encouraged to consume the supplements under supervision 6 d/wk at the local community nutrition center. The number of packets taken by the mothers was recorded during monthly home visits.

Micronutrients

Three types of micronutrients were provided for daily supplementation from week 14 of gestation until delivery; they included MMs, 60 mg Fe (fumarate) + 400 μ g folate (the Fe60 group), and 30 mg Fe (fumarate) + 400 μ g folate (the Fe30 group). The MM group received 15 different vitamins and minerals, developed by the United Nations Children's Fund (UNICEF); 1 tablet contained 150 μ g I (potassium iodide), 15 mg Zn (sulfate), 65 μ g Se (sodium selenite), 2 mg Cu (sulfate), 800 μ g retinyl acetate (RE) vitamin A, 1.4 mg thiamine mononitrate, 1.4 mg vitamin riboflavin, 18 mg vitamin B-3 (niacin), 1.9 mg vitamin B-6 (pyridoxine hydrochloride), 2.6 μ g vitamin B-12 (cyanocobalmin), 70 mg vitamin C, 200 IU vitamin D (vitamin D₃), and 10 μ g vitamin E (α -tocopherol acetate) in the recommended dietary allowance dose in addition to 30 mg Fe (fumarate) and 400 μ g folate. The pills were identical in appearance, and monthly supplies were provided in identical bottles with electronic counting devices (eDEM; AARDEX Inc, Zurich, Switzerland), which had small microprocessors embedded in the caps. The microprocessors recorded the number of times the bottles were opened (27).

Sample

All live singleton babies born from 20 May 2002 to 20 December 2003 ($n = 2853$) constituted our subsample; this number was sufficient to allow detection of a difference of 4 developmental quotient points among the groups with 90% power at the 5% level.

Written informed consent was obtained from the parents or guardians of all of the children. The study was approved by the Ethics Review Committee of ICDDR,B.

Measurements

Socioeconomic status

On enrollment of the mothers, information was collected during a home visit. This information concerned their family wealth (number of possessions—eg, television, radio, domestic animals, chairs, tables, beds, bicycle, or rickshaw), deficits between income and expenditure (occasional or constant—yes or no), family structure and parental characteristics (mother's age, hemoglobin, and parity and parents' education and employment), and housing quality (floor, walls, or roof made of mud—yes or no).

Anthropometry

On enrollment, maternal weight was measured with electronic scales (UNISCALE; Seca GmbH, Hamburg, Germany) that were accurate to 100 g, and height was measured to the nearest 0.1 cm by using a stadiometer. Infant birth weight was measured within 72 h after birth by using beam scales (Seca GmbH), which were precise to 10 g. Research assistants were trained to carry out the anthropometric measurements according to standard procedures (28).

Developmental assessments

When the children were 7 mo old, psychologists who were unaware of the children's groups assessed the children's development in the presence of their mothers at 1 of 4 local clinics.



Two one-step means-end problem-solving tests (PSTs), the support and cover tests, were used to assess cognitive development, and the Psychomotor Developmental Index (PDI) of the Bayley Scales of Infant Development-II was used to assess motor development (29). The original procedures of the PSTs were described by Piaget (30), but the conduct and scoring of the tests were designed by Willatts (31, 32). In these tests, infants manipulate an intermediary to retrieve an object (a toy).

The support test involves placing a long cloth on a table in front of the child and then placing a toy out of the child's reach at the farthest end of the cloth. The infant has to pull the cloth to retrieve the toy. In the cover test, a toy is covered with a cloth while the infant is watching. The infant is then required to remove the cloth to retrieve the toy. These procedures were videotaped and scored later. Four trials were given in both PSTs. In each trial, 3 behaviors—cloth behavior (the way the child handled the cloth), fixation behavior (the way the child fixed his or her vision on the toy), and toy behavior (the way the child grasped the toy)—were scored on a 3-point scale: 0 for no evidence of intention, 1 for possible or ambiguous intention, and 2 for clear evidence of intention. The scores for each behavior were summed to give an intention score for each trial that ranged from 0 to 6, and the scores of 4 trials were then summed to give a total score ranging from 0 to 24. These tests were chosen because they are relatively easy and quick to perform, and because they can be scored from videotape, which facilitates ongoing quality control. The tests have been used in Jamaica (33), where they differentiated LBW from normal-birth-weight infants, were sensitive to an 8-wk intervention of stimulation, and predicted the developmental quotients at the age of 15 mo.

The Bayley Scales of Infant Development-II has been used in Bangladesh by the same group of researchers (34–36); it showed good interobserver and test-retest reliability (36). We also used a modified version of Wolke's behavior rating scale (37) to assess infant behavior during the assessments. This instrument has 5 ratings on 9-point scales that included the following items: the infant's activity level (very still = 1 to overactive = 9), emotional tone (unhappy = 1 to radiates happiness = 9), response to examiner in first 5 min (avoiding = 1 to friendly and inviting = 9), cooperation with the test procedure (resists all suggestions = 1 to always complies = 9), and the amount of vocalization (very quiet = 1 to constant vocalization = 9). These scales also were used previously in Bangladesh (34, 36).

At the beginning of the study, interobserver reliabilities among the 5 testers were assessed on 20 infants. Intraclass correlations for scores of both support and cover PSTs and Bayley PDI were >0.95 , and those for all behavior ratings were >0.89 . Test-retest reliabilities were assessed before the start of the study. Test-retest intraclass correlations over 24 h for both support and cover PST scores of 15 infants were >0.70 and those for the Bayley PDI in 10 infants over a 7-d interval were 0.9. To maintain the quality of testing, ongoing interobserver reliability was assessed weekly on 7% of all tests (PSTs and Bayley PDI: >0.90 ; all behavior ratings: >0.85).

Statistical analysis

We examined differences in enrollment characteristics between the lost and tested samples by using analyses of variance or chi-square tests. We conducted intention-to-treat analyses to assess the intervention effect on all child development outcomes. A 2-factor analysis of covariance (ANCOVA) was conducted by

using micronutrients (3 levels) and food (2 levels) as factors and age as the covariate (where scores differed by age). Scores on the support test, activity, emotion, and cooperation ratings varied by sex, and therefore sex was used as an additional factor for all analyses of these outcomes.

To examine whether thin mothers responded differently to supplements than did better-nourished women, we examined the BMI \times treatment interaction. We divided the sample into infants of mothers with high BMIs and infants of mothers with low BMIs. We took the World Health Organization's definition of BMI [ie, <18.5 for mild undernutrition (38)] as the cutoff for low BMI, and high BMI was ≥ 18.5 . This same value was used by the BINP program to determine which pregnant women to provide with food supplement.

We repeated all ANCOVAs by using maternal BMI (2 levels) as an additional factor. Post hoc Bonferroni tests were conducted to examine any significant difference among the 3 micronutrient groups. When BMI \times treatment interactions were significant, appropriate subgroup analyses were performed. All statistical analyses were performed with the use of SPSS software (version 11.5; SPSS Inc, Chicago, IL).

RESULTS

Loss from the sample

Of the 2853 singleton live births in the subsample, 2116 infants (74%) were tested (**Figure 1**). The main causes for loss to follow-up were being away from home on visits or because of floods (40%); refusal to be tested (27%), probably because of fear of venipuncture; death (16%); moving to a different residence (8%); and being sick at the time of testing (9%). The proportion of lost infants was similar in each of the 6 groups (NS, chi-square test). We first examined any differences between lost and tested children in enrollment characteristics by using a 2-factor analysis of variance (lost or tested by 6 supplement groups). There was no significant difference between lost and tested children in any of the 6 groups in the following characteristics: family income-expenditure deficits; structure of house; mothers' and fathers' educational level; mothers' age, height, and BMI on enrollment; infants' birth weight; and children's age at testing. Overall, mothers of children lost to follow-up had significantly ($P = 0.01$) fewer children (1.13 ± 1.11) than did mothers whose children were tested (1.26 ± 1.1) and also had significantly ($P = 0.02$) higher hemoglobin concentrations at enrollment (11.8 ± 1.3 g/L) than did mothers whose children were tested (11.7 ± 1.3 g/L). Among birth characteristics, infant gestational age was significantly lower in the lost children than in those tested (39.0 ± 2.1 and 39.2 ± 1.7 wk, respectively; $P = 0.03$). However, all differences were small, and none were significantly different across the 6 supplement groups (all 6 lost or tested \times supplement group interactions were NS).

Enrollment characteristics

Enrollment characteristics are shown in **Table 1**. Male infants constituted approximately one-half of the sample. The families were extremely poor; only one-third of the families had ≥ 1 adult in regular employment, and 19% of families had experienced occasional or continuous income-expenditure deficits in the



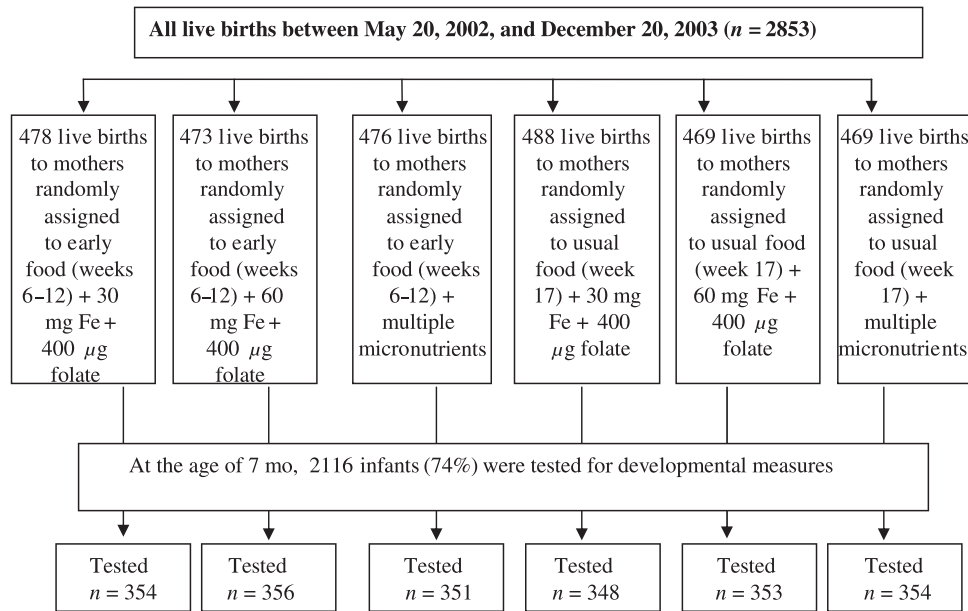


FIGURE 1. Flow chart showing the number of live births in each group during the 19-mo study period and the number of children tested at age 7 mo.

previous year. There was no significant difference among the 6 groups in any measured enrollment or birth characteristics.

Compliance

Compliance was measured up to gestation week 30. The average number of food packets reportedly taken during the first 30 wk of pregnancy was 92 ± 41 in the early-food group and 63 ± 36 in the usual-food group. There was a slight overlap, because 10% of the early-food group began after some of the usual-food group had begun. The number of micronutrient pills taken in the first 30 wk of pregnancy was 79 ± 34 in the Fe30 group, 78 ± 34 in the Fe60 group, and 75 ± 33 in the MM group.

Intent-to-treat analyses

The means (±SDs) of the developmental outcomes are shown in Table 2. Age was significantly positively correlated with support ($r = 0.08, P < 0.001$) and cover ($r = 0.07, P < 0.05$) and significantly negatively correlated with the PDI ($r = -0.21, P < 0.001$). The male infants had significantly better total scores on the support test than did the females (11.5 ± 7.6 and 10.6 ± 7.6 , respectively; $P = 0.02$).

The use of a 2-factor ANCOVA and after control for age (3-factor ANCOVA for support test with sex as a third factor) showed no significant effect of food or micronutrient supplementation on the developmental test scores. We used similar

TABLE 1
Characteristics of the 6 groups at enrollment¹

	Fe30-folate + early food (n = 478)	Fe30-folate + usual food (n = 473)	Fe60-folate + early food (n = 476)	Fe60-folate + usual food (n = 488)	15 MMs + early food (n = 469)	15 MMs + usual food (n = 469)	Total (n = 2853)
Families with ≥1 adult with regular employment (%)	33	36	31	35	38	36	35
Families with income-expenditure deficits (%)	20	19	17	19	20	19	19
Families having a house with mud construction (%)	22	20	20	18	25	21	21
Fathers with no education (%)	30	31	29	33	33	28	31
Mothers with no education (%)	34	31	32	33	34	32	33
Mothers with BMI (kg/m ²) < 18.5 (%)	29	32	29	30	27	24	28
Primipara (%)	35	34	35	34	30	35	34
Mother's age (y)	26 ± 6 ²	26 ± 6	26 ± 6	26 ± 6	27 ± 6	26 ± 6	26 ± 6
Mother's BMI at enrollment	20.2 ± 2.6	19.9 ± 2.8	20.1 ± 2.7	20 ± 2.7	20.2 ± 2.6	20.5 ± 2.5	20 ± 3
Maternal hemoglobin (g/L)	117 ± 13	117 ± 12	116 ± 13	116 ± 13	117 ± 13	118 ± 13	117 ± 13
Male/female (n)	196/158	176/175	174/179	173/183	185/163	179/175	—

¹ Fe30, 30 mg Fe/d; Fe60, 60 mg Fe/d; MMs, multiple micronutrients. There was no significant difference among the 6 groups in any enrollment measures (chi-square test for percentages and ANOVA for means).

² $\bar{x} \pm SD$ (all such values).

TABLE 2Total problem-solving test (PST), motor development, and behavior scores of infants in 6 treatment groups¹

	Early-food group (n = 1058)			Usual-food group (n = 1058)			Significant findings ²
	Fe30	Fe60	MM	Fe30	Fe60	MM	
Age at testing (d)	223 ± 8	223 ± 8	223 ± 8	222 ± 7	223 ± 9	223 ± 9	
Male//female (n)	196/158	176/175	174/179	173/183	185/163	179/175	
PST							
Support							
H	11.2 ± 7.4	11.2 ± 7.6	10.8 ± 7.7	11.8 ± 7.3	10.9 ± 7.4	11.8 ± 7.6	BMI, <i>P</i> = 0.018
L	10.8 ± 7.6	10.5 ± 8	11.6 ± 8.1	9 ± 7.7	9.2 ± 7.6	11.2 ± 8.1	Sex, <i>P</i> = 0.03 BMI × food, <i>P</i> = 0.024
Cover							
H	13.1 ± 6.9	12.6 ± 7.2	12.7 ± 7.1	13.2 ± 6.6	13.6 ± 7	13.3 ± 7	BMI, <i>P</i> = 0.039
L	11.9 ± 6.9	12.5 ± 7.1	13.8 ± 6.8	12.1 ± 7.4	11.7 ± 7.3	12.3 ± 7.3	BMI × food, <i>P</i> = 0.058
Motor development							
Bayley Scale (PDI)							
H	103.6 ± 16.1	104 ± 14.7	102.8 ± 16.2	103.2 ± 15.4	103.5 ± 15.3	104.3 ± 16.4	BMI, <i>P</i> = 0.003
L	100.9 ± 15.6	101.9 ± 15.4	102.6 ± 17.3	98.4 ± 14.3	99.3 ± 15.1	105.3 ± 16.4	Mn, <i>P</i> = 0.023 BMI × Mn, <i>P</i> = 0.05
Behavior							
Approach							
H	5.9 ± 1.1	6 ± 1.1	5.8 ± 1.2	5.8 ± 1.2	5.9 ± 1.2	5.8 ± 1.2	—
L	5.9 ± 1.4	5.9 ± 1.3	5.9 ± 1.2	5.7 ± 1.4	5.8 ± 1.2	5.9 ± 1.3	
Activity							
H	4.8 ± 1.3	4.9 ± 1.4	4.7 ± 1.4	4.9 ± 1.3	4.8 ± 1.4	4.9 ± 1.4	BMI, <i>P</i> = 0.004
L	4.6 ± 1.4	4.6 ± 1.4	4.8 ± 1.3	4.4 ± 1.2	4.7 ± 1.4	4.8 ± 1.3	Sex, <i>P</i> = 0.038 BMI × Mn, <i>P</i> = 0.048
Vocalization							
H	3 ± 1.6	3.1 ± 1.7	2.9 ± 1.7	3.3 ± 1.6	3 ± 1.9	3.2 ± 1.7	BMI, <i>P</i> = 0.017
L	2.9 ± 1.7	3.1 ± 1.8	2.9 ± 1.6	2.7 ± 1.8	2.9 ± 1.5	2.8 ± 1.7	BMI × food, <i>P</i> = 0.036
Emotion							
H	5 ± 1.4	4.9 ± 1.5	4.7 ± 1.6	5.1 ± 1.5	4.9 ± 1.6	5 ± 1.5	BMI, <i>P</i> = 0.028
L	4.8 ± 1.7	4.9 ± 1.6	4.9 ± 1.5	4.5 ± 1.6	4.6 ± 1.7	4.9 ± 1.6	Sex, <i>P</i> = 0.011 BMI × food, <i>P</i> = 0.039
Cooperation							
H	5.1 ± 1.5	5 ± 1.6	4.8 ± 1.6	5.1 ± 1.4	5 ± 1.5	5 ± 1.5	BMI × food, <i>P</i> = 0.037
L	4.9 ± 1.7	5 ± 1.7	5 ± 1.6	4.6 ± 1.6	4.7 ± 1.7	5 ± 1.6	Sex, <i>P</i> = 0.002

¹ All values are $\bar{x} \pm$ SD. Fe30, 30 mg Fe/d; Fe60, 60 mg Fe/d; MM, multiple micronutrients (containing 15 vitamins and minerals); H, high-BMI group; L, low-BMI group; Mn, 3 micronutrient groups.

² ANCOVA with age as covariate and mothers' BMI, food group, and micronutrient group as factors (also sex in analyses of support and activity rating). All 3-way interactions and interactions between the treatments were NS.

ANCOVAs with the 5 behavior ratings (approach, activity, emotional tone, vocalization, and cooperation). Age was significantly ($P < 0.01$) correlated with cooperation and was used as a covariate for that analysis. Male children had significantly higher ratings than females for activity ($P < 0.05$) and cooperation and emotional tone ($P < 0.01$ for both); therefore, sex was an extra factor in analyses of those behaviors. There was no significant effect of supplementation on any of the behavior ratings.

Effect of supplementation stratified on mothers' BMI in early pregnancy

We had initially hypothesized that supplementation would benefit undernourished pregnant women more than better-nourished pregnant women. Accordingly, women were divided into 2 groups according to their BMI on enrollment at 8–10 wk of pregnancy—the better-nourished (BMI ≥ 18.5) and the undernourished (BMI < 18.5) groups—and a second series of analyses of variance was conducted with the addition of BMI group as a factor (Table 2).

Problem-solving tests

Maternal BMI had a significant effect on support ($P = 0.018$) and cover ($P = 0.039$) test scores. In both tests, the infants of undernourished mothers had slightly but significantly lower scores than did the infants of better-nourished mothers [support test: 10.25 ± 7.85 and 11.27 ± 7.49 , respectively ($P = 0.006$); cover test: 12.31 ± 7.14 and 13.10 ± 6.97 , respectively ($P = 0.022$)]. In the support test, the BMI × food group interaction was significant ($P = 0.024$), and it approached significance in the cover test ($P = 0.058$). The 3-way interaction of the 2 groups of treatment was not significant in either problem-solving test. The mean scores of the support test in children of low-BMI mothers in the early- and usual-food groups were 10.9 ± 7.9 and 9.6 ± 7.8 , respectively, which gave an effect size (z score) of 0.17 (95% CI: 0.01, 0.33; $P = 0.035$). In contrast, in the children of the high-BMI mothers, the early- and usual-food group mean scores were 11.06 ± 7.53 and 11.49 ± 7.45 , respectively, which gave an effect size (z score) of -0.06 (95% CI: $-0.16, 0.04$; $P = 0.25$).



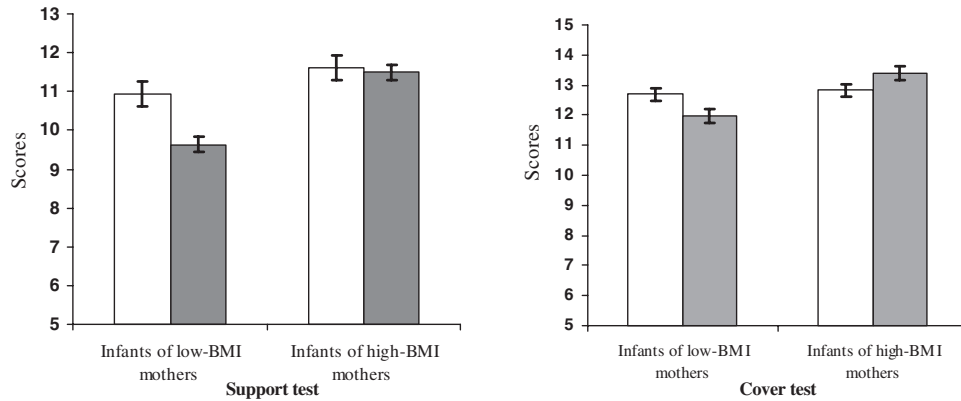


FIGURE 2. Mean (\pm SE) scores on problem-solving support and cover tests for infants of mothers with low or high BMI by early- (□) or usual- (■) food group. Early-food group: low BMI, $n = 278$; high BMI, $n = 777$. Usual-food group: low BMI, $n = 309$; high BMI, $n = 742$. Support test: food \times BMI interaction, $P = 0.024$; low BMI early food versus usual food, $P = 0.035$; high BMI early food versus usual food, NS (ANOVA). Cover test: food \times BMI interaction, $P = 0.058$ (ANOVA).

(Figure 2). There was no significant BMI \times micronutrient interaction in any of the PSTs.

The Psychomotor Developmental Index

Maternal BMI also had a significant ($P = 0.003$) effect on the PDI: infants of undernourished mothers had slightly lower scores (101.06 ± 15.66) than did infants of better-nourished mothers (103.52 ± 15.68). With BMI in the model, micronutrient group had a significant effect, but the micronutrient \times BMI interaction was significant ($P = 0.05$), and the interaction of the 2 treatment groups with BMI was not significant. In infants of the low-BMI mothers, the means of the MM and Fe30 groups were 103.9 ± 16.87 and 99.5 ± 14.88 , respectively, which gave an effect size (z score) of 0.28 (95% CI: 0.08, 0.48; $P < 0.01$). The mean of the Fe60 group was 100.49 ± 15.27 , for a difference (z score) of 0.22 (95% CI: 0.01, 0.42; $P = 0.04$) from the MM group's score (Figure 3). In contrast, there was no effect in the high-BMI group. The interactions of food with BMI or with food and micronutrients were not significant.

Effects on behavior ratings

BMI also had significant effects on activity ($P = 0.004$), emotion ($P = 0.029$), and vocalization ($P = 0.018$) ratings. Infants of

better-nourished mothers tended to be more active and happier and to vocalize more during the test than did infants of undernourished mothers. Neither food nor micronutrients had significant effects in any behavior. The interaction between BMI and food was significant in analyses of vocalization ($P = 0.04$), emotion ($P = 0.04$) and cooperation ($P = 0.04$). In each case, infants of low-BMI mothers tended to benefit from early food, whereas those of high-BMI mothers did not; however, the benefit was extremely small and not significant. The BMI \times micronutrient group interaction was significant ($P = 0.048$) in analysis of activity. In the low-BMI groups, the mean scores of the MM, Fe60, and Fe30 groups were 4.78 ± 1.33 , 4.63 ± 1.41 , and 4.45 ± 1.29 , respectively, which gave an effect size (z score) of 0.24 (95% CI: 0.037, 0.45; $P = 0.018$) between the MM and Fe30 groups. There was no significant food \times micronutrients interaction for any of the behavioral variables, and none of the interactions of the 2 treatments with BMI were significant.

To determine whether grouping the children according to the mother's BMI status (low or high) had introduced confounding, we compared the characteristics of the treatment groups in low-BMI mothers only. There were no significant differences among the groups in any of the enrollment characteristics shown in Table 1 or in the age and sex of the children.

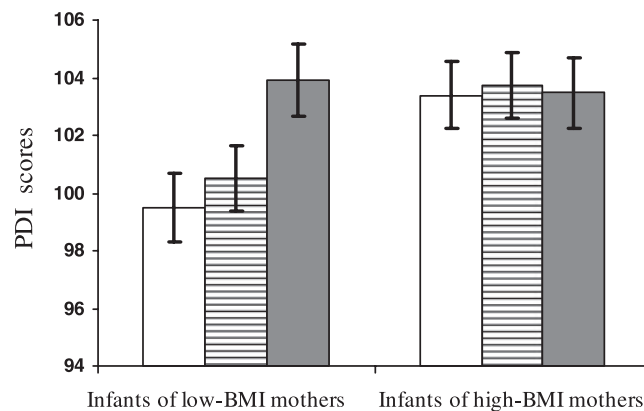


FIGURE 3. Mean (\pm SE) Psychomotor Developmental Index (PDI) values for infants of mothers with low or high BMI by micronutrient group. □, 30 mg Fe + folate; ▨, 60 mg Fe + folate (F); ■, 15 multiple micronutrients (MMs). 30 mg Fe+F group: low BMI, $n = 216$; high BMI, $n = 491$; 60 mg Fe+ F group: low BMI, $n = 210$; high BMI, $n = 486$; MM group: low BMI, $n = 161$; high BMI, $n = 540$. MMs \times BMI interaction, $P = 0.05$, low BMI: MMs versus 30 mg Fe+F, $P = 0.01$, MM versus 60 mg Fe+ F, $P = 0.04$; high BMI: all NS (ANOVA for all).

DISCUSSION

Supplementation in pregnancy with either early or usual food, with MMs, or with either of the iron and folate mixtures provided no overall benefit for child development. However, in infants of low-BMI mothers, early-food supplementation had slightly but significantly greater benefits than did usual-food supplementation. MM supplementation during pregnancy also had a somewhat greater benefit for infant motor development and activity levels than did the 2 combinations of iron-folate supplementation. In contrast, we found no such benefits in infants of higher-BMI mothers. This study is the only one of which we are aware that has examined the effect of different durations of food supplementation on children's development.

The present trial was a large, randomized, controlled trial; the testers were unaware of children's groups; the mothers were unaware of their micronutrient supplement; and the tests were reliable. The loss of subjects was not biased, and we had initially hypothesized that low-BMI mothers were more likely than higher-BMI mothers to benefit from supplementation. It is therefore likely that the benefits can be attributed to the treatments. The population was a highly suitable one in which to test the effect of supplementation, because many of the mothers were undernourished and the children had a small average birth weight and a high incidence of LBW.

Food supplementation

The average time difference in commencing food supplements between the early- and usual-food groups was 31 d, and 10% of the early-food group overlapped with some of the usual-food group in starting time. The amount of food supplements reportedly taken by the early- and usual-food groups differed by a total of 18 600 kcal (77 875 kJ). However, it is possible that the amount actually consumed by the mothers was less and that the food was replaced or shared with other family members in these poor households (39). We cannot be certain whether differences in timing or total amount received (or in both) were the important factor. It is surprising that this relatively small difference in supplement had an effect on infants' development. Had there been a group not receiving supplement, it is likely that the difference would have been considerably greater. To establish the full value of food supplementation, the need remains to examine its effects, when given to undernourished women throughout pregnancy, on the development of their children.

In Taiwan, food supplementation throughout pregnancy benefited infants' motor but not mental development at the age of 8 mo more than did a placebo (23). However, the investigators did not analyze the benefits according to the mothers' BMI. They used the Bayley Scales to assess mental development, and it is possible that the PSTs that we used are more sensitive to subtle changes than are the Bayley Scales.

Micronutrient supplementation

The motor development of infants of undernourished mothers also benefited from MMs, whereas that of infants of better-nourished mothers did not. Mothers with low BMIs also may be deficient in MMs. In a previous trial in the same community, infants supplemented with MMs showed greater improvements in motor scores than did infants receiving riboflavin (35). In the present study, in addition to micronutrients, all mothers received food supplementation, and, therefore, we should be cautious

before extrapolating the findings to undernourished populations to whom food supplementation is not available.

It is possible that more subtle changes to the brain, such as improvement in neurotransmitters, dendritic branching, connections, and myelination, may have occurred (40, 41). However, the size of the benefit in development from both MMs and early-food supplementation is very small, and it is not certain whether there will be any functional advantage.

The consistent association of mothers' BMI with both cognitive and motor function provides further evidence of the importance of maternal nutrition and child development. From a policy perspective, it appears that food supplementation should begin as early as possible in pregnancy and that it should be targeted toward undernourished women; moreover, efforts should be made to improve compliance. In addition, in countries where there are no food supplementation programs and where large numbers of women are undernourished, it is likely that the offspring are at high risk of poor development. In contrast to food supplementation, benefits of prenatal MM supplementation on birth outcomes are less clear; this is the first study that we know of that examined the effects on child development; and the benefits were limited to motor development and were small. Longer-term follow-up is required to determine whether any of the observed benefits have functional importance.

We thank Peter Willatts for training us on the problem-solving tests and guiding us in testing the children and the mothers and families in Matlab who participated in this study. We also thank all our testers and supervisors, who made tremendous efforts to maintain the quality of the information collected.

The authors' responsibilities were as follows—LAP and SEA: the conduct of the main study; E-CE: designing and implementing the micronutrient component; SMG-M, FT, JDH, and SNH: writing the proposal, designing the child development portion of the study, supervision of the conduct of the study, and data analysis; FT, JDH, SNH, and FM: subject recruitment, data collection, and logistical organization of the child development portion of the study; DR: statistical analysis; and all authors: review and critique of the manuscript drafts.. This report is a part of FT's doctoral research work. None of the authors had a personal or financial conflict of interest.

REFERENCES

1. State of the world's children 2006. Internet: www.unicef.org/bangladesh/Child_and_Maternal_Nutrition.pdf (accessed 28 January 2007).
2. Jiang T, Christian P, Khatry SK, Wu L, West KP Jr. Micronutrient deficiencies in early pregnancy are common, concurrent, and vary by season among rural Nepali pregnant women. *J Nutr* 2005;135(5):1106–12.
3. UNICEF/UNU/WHO. Composition of multiple micronutrient supplement to be used in pilot programmes among pregnant women in developing countries. New York, NY: UNICEF, 1999.
4. UNICEF/UNU/WHO. Multiple micronutrient supplementation during pregnancy (MMSDP): efficacy trials. London, United Kingdom: Centre for International Child Health, Institute of Child Health, 2002:1–21.
5. World Health Organization. Iron deficiency anaemia: assessment, prevention and control. A guide for programme managers. Geneva, Switzerland, World Health Organization, 2001. (WHO/NHD/01.3.)
6. Kramer MS, Kakuma R; Energy and protein intake in pregnancy. *Cochrane Database Syst Rev* 2003;(4):CD000032.
7. Osrin D, Vaidya A, Shrestha Y, Baniya RB, Manandhar DS, Adhikari RK, Filteau S, Tomkins A, Costello AM; Effects of antenatal multiple micronutrient supplementation on birthweight and gestational duration in Nepal: double-blind, randomised controlled trial. *Lancet* 2005;365(9463):955–62.
8. Hininger I, Favier M, Arnaud J, et al. Effects of a combined micronutrient supplementation on maternal biological status and newborn anthropometrics measurements: a randomized double-blind, placebo-controlled trial in apparently healthy pregnant women. *Eur J Clin Nutr* 2004;58(1):52–9.



9. Christian P, Shrestha J, LeClerq SC, et al. Supplementation with micronutrients in addition to iron and folic acid does not further improve the hematologic status of pregnant women in rural Nepal. *Nutrition* 2003; 133(11):3492–8.
10. Friis H, Gomo E, Nyazema N, et al. Effect of multimicronutrient supplementation on gestational length and birth size: a randomized, placebo-controlled, double-blind effectiveness trial in Zimbabwe. *Am J Clin Nutr* 2004;80(1):178–84.
11. Ramakrishnan U, Gonzalez-Cossio T, Neufeld LM, Rivera J, Martorell R. Multiple micronutrient supplementation during pregnancy does not lead to greater infant birth size than does iron-only supplementation: a randomized controlled trial in a semirural community in Mexico. *Am J Clin Nutr* 2003;77(3):720–5.
12. Susser E, Neugebauer R, Hoek HW, et al. Schizophrenia after prenatal famine. Further evidence. *Arch Gen Psychiatry* 1996;53:25–31.
13. Kirksey A, Rahmanifar A, Wachs TD, et al. Determinants of pregnancy outcome and newborn behavior of a semirural Egyptian population. *Am J Clin Nutr* 1991;54(4):657–67.
14. Chavez A, Martinez H, Guarneros N, Allen L, Pelto G. [Nutrition and development in the first semester of life.] *Salud Publ Mex* 1998;40(2): 111–8 (in Spanish).
15. Dobo M, Czeizel AE; Long-term somatic and mental development of children after periconceptional multivitamin supplementation. *Eur J Pediatr* 1998;157(9):719–23.
16. Botto LD, Mulinare J, Erickson JD. Occurrence of omphalocele in relation to maternal multivitamin use: a population-based study. *Pediatrics* 2002;109(5):904–8.
17. Merialdi M, Caulfield LE, Zavaleta N, Figueroa A, DiPietro. Adding zinc to prenatal iron and folate tablets improves fetal neurobehavioral development. *Am J Obstet Gynecol* 1999;180(2 Pt 1):483–90.
18. Levitsky DA, Strupp BJ. Malnutrition and the brain: changing concepts, changing concerns. *J Nutr* 1995;125(suppl):2212S–20S.
19. Pencharz P, Heller A, Higgins A, Strawbridge J, Rush D, Pless B. Effect of nutritional services to pregnant mothers on the school performance of treated and untreated children. *Nutr Res* 1983;3:795–803.
20. Osofsky HJ. Relationships between prenatal medical and nutritional measures, pregnancy outcome, and early infant development in an urban poverty setting. I. The role of nutritional intake. *Am J Obstet Gynecol* 1975;123(7):682–90.
21. Rush D, Stein Z, Susser M. A randomized controlled trial of prenatal nutritional supplementation in New York City. *Pediatrics* 1980;65(4): 683–97.
22. Joos SK, Pollitt E, Mueller WH, Albright DL. The Bacon Chow study: maternal nutritional supplementation and infant behavioral development. *Child Dev* 1983;54(3):669–76.
23. Hsueh AM, Meyer B. Maternal dietary supplementation and 5 year old Stanford Binet IQ test of the offspring in Taiwan. *Fed Proc* 1981;40:897.
24. Waber DP, Vuori-Christiansen L, Ortiz N, et al. Nutritional supplementation, maternal education, and cognitive development of infants at risk of malnutrition. *Am J Clin Nutr* 1981;34(suppl):807–13.
25. Grantham-McGregor SM, Ani CC. The role of micronutrients in psychomotor and cognitive development. *Br Med Bull* 1999;55(3):511–27.
26. Czeizel AE, Dobo M. Postnatal somatic and mental development after periconceptional multivitamin supplement. *Arch Dis Child* 1994;70(3): 229–33.
27. Ekström, E-C, Hyder Z, Chowdhury AMR, et al. Efficacy and trial effectiveness of weekly and daily iron supplementation among pregnant women in rural Bangladesh: disentangling the issues. *Am J Clin Nutr* 2002;76:1392–400.
28. World Health Organization. Measuring change in nutritional status. Guidelines for assessing the nutritional impact of supplementary feeding programmes for vulnerable groups. Geneva, Switzerland: World Health Organization, 1983.
29. Bayley N. The Bayley scales of infant development. New York, NY: Psychological Corporation, 1970.
30. Piaget J. The construction of reality in the child. London, United Kingdom: Routledge and Kegan Paul, 1955.
31. Willatts P. Stages in development of intentional search by young infants. *Dev Psychol* 1984;20(3):389–96.
32. Willatts P. Development of means-end behavior in young infants: pulling a support to retrieve a distant object. *Dev Psychol* 1999;35(3):651–67.
33. Meeks Gardner J, Walker SP, Powell CA, Grantham-McGregor S. A randomized controlled trial of a home-visiting intervention on cognition and behavior in term low birth weight infants. *J Pediatr* 2003;143(5): 634–9.
34. Tofail F, Kabir I, Hamadani JD, et al. Supplementation of fish-oil and soy-oil during pregnancy and psychomotor development of infants. *J Health Popul Nutr* 2006;24(1):48–56.
35. Black MM, Baqui AH, Zaman K, et al. Iron and zinc supplementation promote motor development and exploratory behavior among Bangladeshi infants. *Am J Clin Nutr* 2004;80(4):903–10.
36. Hamadani JD, Fuchs GJ, Osendarp SJ, Huda SN, Grantham-McGregor SM. Zinc supplementation during pregnancy and effects on mental development and behaviour of infants: a follow-up study. *Lancet* 2002; 360(9329):290–4.
37. Wolke D, Skuse D, Mathisen V. Behavioral style in failure to thrive infants: a preliminary communication. *J Pediatr Psychol* 1990;15:237–54.
38. World Health Organization. Global database on body mass index. Internet: http://www.who.int/bmi/index.jsp?introPage=intro_3.html (last updated 10 September 2007).
39. Abdullah M, Karim R, Samad QA. Is the supplement a substitute or true supplementation to the usual diet? Results from the Bangladesh Integrated Nutrition Project (BINP) Operations Research project (ORP). Dhaka, Bangladesh: International Centre for Diarrhoeal Disease Research, 2000:6–7.
40. Chase HP, Welch NN, Dabiere CS, Vasan NS, Butterfield LJ. Alterations in human brain biochemistry following intrauterine growth retardation. *Pediatrics* 1972;50(3):403–11.
41. Johnson MH. Developmental cognitive neuroscience. 2nd ed. Melbourne, Australia: Blackwell Publishing, 2005.