

The MAL-ED Cohort Study in Mirpur, Bangladesh

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The Etiology, Risk Factors and Interactions of Enteric Infections and Malnutrition and the Consequences for Child Health and Development (MAL-ED) study site in Bangladesh is located in the capital city of Dhaka in an urban slum that has one of the highest population densities in the world. The site is in the Bauniabadh area of Mirpur, Dhaka. A typical squatter settlement, the average family size of households in Mirpur Bauniabadh is 4.5, with 48% females. About 20% of households have a monthly income of only US\$62. About 30% of mothers never attended school, and only 3% obtained secondary school education. The majority of the people are day laborers, garment workers, and transport workers. About 72% of caregivers always wash their hands after helping the child defecate and 6.6% never wash their hands. The diarrheal attack rate for Mirpur is 4.69 episodes per child per year. The study site is representative of a typical urban slum of Dhaka city in terms of demographics, socioeconomic status, and general health indicators.

Keywords. Bangladesh; birth cohort; case-control; MAL-ED; malnutrition.

The Etiology, Risk Factors and Interactions of Enteric Infections and Malnutrition and the Consequences for Child Health and Development (MAL-ED) network is conducting a multicountry, longitudinal prospective cohort study on the etiology, risk factors, and interactions of enteric infections and malnutrition, and the consequences of these factors on child growth, cognitive development, and vaccine response. The 8 study sites are epidemiologically and geographically diverse and comprised of low-income populations. The 8 MAL-ED cohort sites are located in Bangladesh, Brazil, India, Nepal, Pakistan, Peru, South Africa, and Tanzania.

The rate of childhood malnutrition has decreased in Bangladesh over the last several decades; however, it unfortunately remains one of the highest in the world [1]. The MAL-ED study in Bangladesh (BGD) is being conducted among residents of an underprivileged

community in Mirpur, one of the 21 administrative units of the nation's capital, Dhaka. Mirpur was selected as the study site because it is inhabited by poor and middle-class families, residential and sanitary conditions are typical of any congested urban settlement, and the study investigators have ongoing field research activities in the area. Mirpur has a population of about half a million in an area of 14.22 km² [1]. It is divided into several sections, and the MAL-ED study is being conducted in the Bauniabadh area of section 11 of Mirpur (Figure 1).

In Bangladesh, icddr,b (formerly the International Centre for Diarrhoeal Disease Research, Bangladesh) is the MAL-ED implementing partner with the University of Virginia. The BGD study area is densely populated and is located 7–8 km from the icddr,b Dhaka Hospital at Mohakhali. There is a history of conducting research studies in BGD by icddr,b staff. A primary healthcare clinic is operated in the BGD study area by icddr,b. All community-based research studies are supported by services available from the icddr,b. These services include a hospital internationally known for treatment of gastrointestinal infections, severe acute malnutrition, acute respiratory infections, and bloodstream infections. Additional services include sophisticated laboratories in

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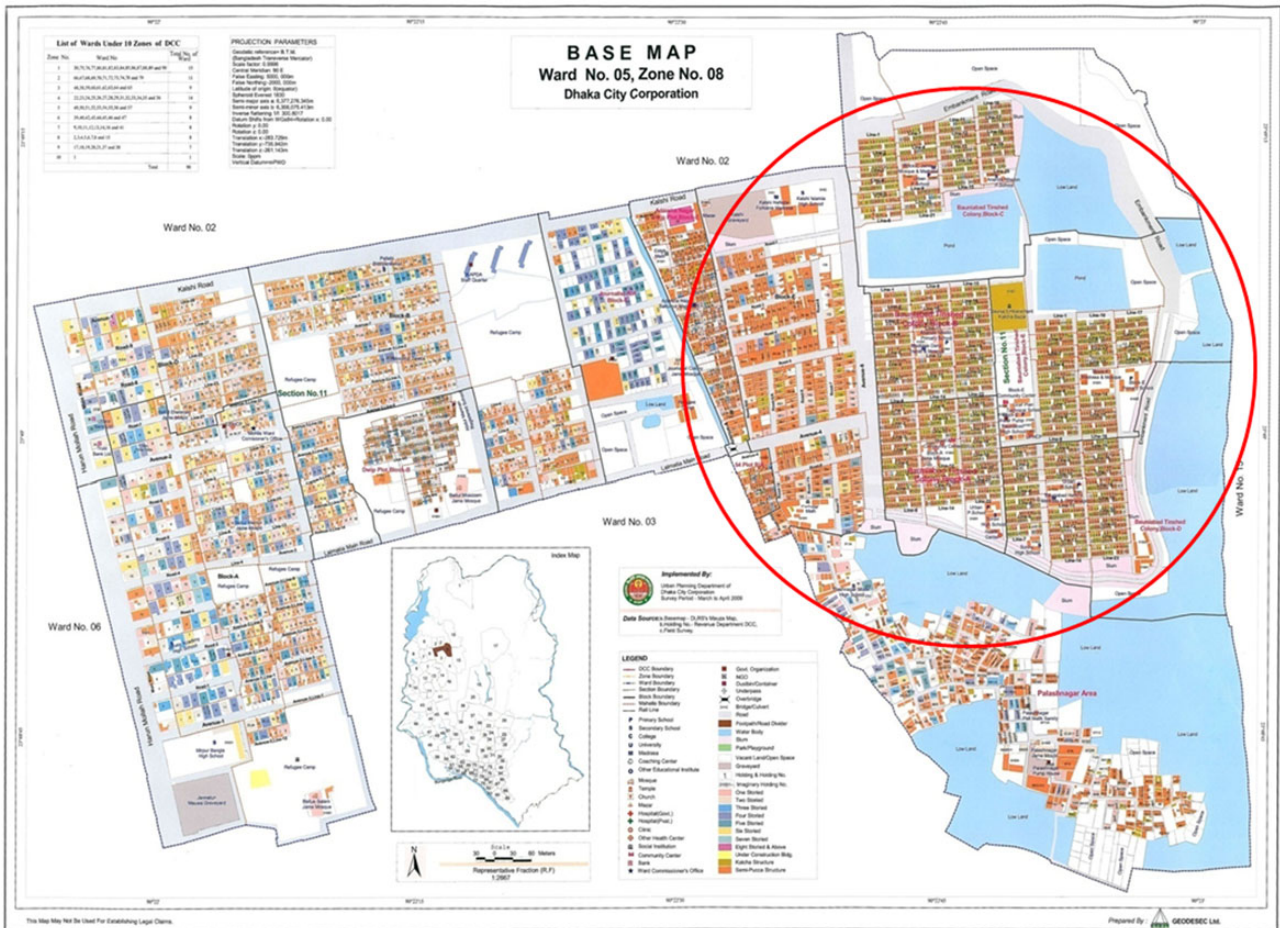


Figure 1. MAL-ED Study Site, Mirpur, Dhaka, Bangladesh. The figure shows the geographic location of the MAL-ED study site of Bangladesh in Dhaka City Corporation. Additionally, figure illustrates the map of squatter settlement of study area situated in Bauniabadh slum, Mirpur, Dhaka. Source: Urban planning department, Dhaka City Corporation North. Available at: <http://www.dncc.gov.bd/departments-with-function/mayor-other/urban-planning.html>.

microbiology, molecular biology, nutritional biochemistry, immunology, and parasitology and experts in epidemiology, biostatistics, clinical medicine, and health systems.

Climate

Mirpur, Dhaka has a hot and humid tropical climate and belongs to the tropical savanna climate [2]. Bangladesh has 3 distinct seasons: the pre-monsoon hot season from March through May; the rainy monsoon, which lasts from June through October; and a cool, dry winter from November through February. The annual average temperature is 25°C (71°F), and it varies from 18°C (64.4°F) in January to 32°C (90°F) in May. But, in recent years, winter temperatures have dropped to around 10°C. The average annual rainfall for Dhaka city is 2123 mm [3].

Population of Mirpur

Initially, the majority of the inhabitants were of Bihari ethnic origin, who settled in Mirpur long before the war with Pakistan

in 1971; however, intensive cultural and social integration has occurred since then and now the population is ethnically diverse. The population of the BGD study area is approximately 87 000, and is stable with low socioeconomic conditions and suboptimal sanitation [4].

Comparison of Site-Specific and National Data

The socioeconomic and demographic data of the BGD site was compared with national, national urban, and Dhaka city data. Site-specific data from published sources were not available for all parameters (Table 1). The population density of the BGD site is one of the highest in Dhaka city. More than 38 000 people live in each square kilometer of the BGD site compared with the mean of 8229/km² in Dhaka district and 976/km² in Bangladesh. However, the family size and sex distribution was comparable to national data. The family size is 4.5 in BGD and 4.21 for Dhaka city [4]. There were differences in the major employment categories—more people in the BGD site were day laborers,

Table 1. Site-Specific and National Data on Health and Demographics for the Dhaka, Bangladesh, MAL-ED Site

Feature	BGD Study Site	National, Regional, or Dhaka City
Population density	38 760/km ² (Mirpur) [5]	8229/km ² (Dhaka city) [4]
Population in catchment area	111 811 [4]	12 043 977 (Dhaka city) [4]
Female sex	48% [4]	46% [4]
Average family size	4.5 [4]	4.2 [6]
Life expectancy, y	NA	M: 68.2, F: 69.2 [7]
Major employment	Day laborer, garment worker, transport worker, small business	Agriculture, small business, transport and unskilled worker [6]
Access to clean water	93% [7]	97% [6]
Access to electricity	100%	47%
Access to improved toilet/sanitation	76%	30% [6]
Women deliver at home	NA	52% (urban Bangladesh) [8]
Ante- and postnatal care	NA	Antenatal: 83% Postnatal: 27% [8] (urban Bangladesh)
Under-5 mortality rate per 1000 live births	NA	53 [8]
Diarrhea burden	4.69 episodes per child per year [9]	NA
HIV prevalence	NA	<0.1 (15–49 y) [10]
Tuberculosis rate per 100 000	NA	Prevalence: 387 Incidence: 223 [11]
Per capita GDP, US\$	NA	755 [4]

Abbreviations: GDP, Gross Domestic Product; HIV, human immunodeficiency virus; NA, not available.

garment workers, and transport workers, whereas agriculture, business, and unskilled workers are the 3 major employment categories in Bangladesh. About 47% of households have electricity at the national level, compared with 100% in the study site. Improved water supply was comparable between the site-specific and national data, but whereas 75% of households in the BGD site use improved toilets, only 30% of households in urban Bangladesh have similar facilities (Table 1).

Socioeconomic Status Within the BGD MAL-ED Study Site

A pilot socioeconomic status (SES) survey was conducted in the MAL-ED study site catchment area before starting the cohort study [12]. The BGD site has 5 blocks (labeled A–E). Each block has an average of 1200 households. For the pilot survey, 20 mothers were selected randomly from each block for interview (total $n = 100$). After completion of the pilot SES survey, the SES survey was repeated with mothers from within the MAL-ED BGD study cohort at 6, 12, 18, and 24 months of follow-up.

Mothers in the pilot SES study were 3 years older than the cohort mothers, and their ages at first pregnancy were 18.4 years and 18.7 years, respectively (Table 2). The cohort mothers had been living in the community longer than the pilot mothers. Seventy-five percent of the families of pilot mothers and 65% of families of cohort mothers had only 1 room for sleeping. On average, 4.8 persons slept in the households of the pilot study and 5.2 persons in the households of the cohort study. The main household building materials were comparable between the groups. Most of the roofs of households are made of metal or

iron sheets, and the floor was made of cement or concrete. About 57% of mothers in the pilot study—compared with 64% in the cohort—treated water by some means to make it safer for drinking; a majority treated it by boiling. Around half of the households cooked inside the house, with only 7%–10% households in both groups having a separate kitchen. A total of 72% of cohort mothers reported washing their hands after cleaning the child following defecation, whereas this was stated by only 48% of mothers from the pilot study. More mothers in the pilot study (32%) vs cohort study (19%) reported receiving no schooling. However, 27% of mothers in the pilot study, and 33% in the cohort study reported completing primary school. The mean monthly income of the cohort households was US\$125, whereas it was US\$113 for pilot households.

The household asset index was constructed using household asset data obtained from the SES questionnaires employed in the pilot and the cohort study populations. From these asset-related dichotomous variables, principal components analysis was performed to produce a common factor score for each household. After ranking by their score, and then dividing the ranking into 3 equal parts, low (poor), intermediate, and high (wealthy) households were identified. The household asset indices were nearly identical between the 2 groups surveyed. However, household food security access status by using the Food and Nutrition Technical Assistance project's Household Food Insecurity Access Scale showed that only 34% of households from the pilot study compared with 72% of households from the cohort study had food security (Table 2).

Table 2. Socioeconomic Data from Pilot and Cohort Study Socioeconomic Status Survey

Characteristic	Pilot SES Survey (n = 100)	SES Survey of Cohort (n = 241)
Age of mother, y	27.64 ± 6.12 (19–45)	24.92 ± 4.98 (17–40)
Age at first pregnancy, y	18.40 ± 2.72	18.74 ± 3.07
No. of live births	2.6 ± 1.76	1.96 ± 1.12
Duration family has lived in house, y	2.47 ± 1.38 y	2.8 ± 1.39 y
No. of room(s) in the household	1.48 ± 0.89 (1–5)	1.62 ± 0.96 (1–6)
No. of rooms for sleeping	1.35 ± 0.67 (1–4)	1.53 ± 0.78 (1–5)
No. of people who usually sleep in the household	4.8 ± 1.95 (2–15)	5.19 ± 2.31 (1–14)
Main building materials for house (roof), %		
Metal	87	87
Wood	2	12
Cement or concrete	11	0.8
Main building materials for house (floor), %		
Earth or sand	11	6
Wood	1	1
Ceramic	1	0.4
Cement or concrete	87	92
Main building materials for house (wall), %		
Bamboo or cane	3	3
Cement or concrete	12	12
Bricks	39	39
Metal	45	45
Other	1	1
Separate room for kitchen, %	10	8
Cooking done, %		
Inside house	48	54
Outside house	50	45
Both inside and outside house	2	0.4
Type of cooking stove use, %		
Gas stove	73	79
Open fire	20	11
Electric heater	6	9.7
Kerosene stove	0	0.4
Other	1	1
Main source of drinking water, %		
Piped into dwelling	9	19
Piped to yard and plot	89	81
Public tap	2	0.4
Treat water to make it safe, %	57	64.3*
Water treatment at household, %		
Boil	74	90*
Chlorine or bleaching powder	1.7	1

Table 2 continued.

Characteristic	Pilot SES Survey (n = 100)	SES Survey of Cohort (n = 241)
Water filter	1.7	2
Let it stand and settle	3.4	2
Other	19	5*
Wash hands after helping child defecate, %		
Always	48	73*
Sometimes	42*	14
Rarely	5	7
Never	5	7
Education level of mother, %		
No schooling	32	19
Primary incomplete (1–5 y)	39	47
Primary completed (6–10 y)	27	33
Secondary completed or higher	3	1
Monthly household income, %		
≤5000 BDT (US\$62)	24	21
5001–10 000 BDT (US\$62–\$123)	58	54
10 001–15 000 BDT (US\$123–\$185)	11	14
>15 000 BDT (>US \$185):	7	11
Household asset index, %		
Low	34	32
Intermediate	33	34
High	33	33
Household food security access, %		
Food secure	34	73
Mildly food insecure	15	6
Moderate food insecure	33	13
Severe food insecure	18	9

Data are presented as mean±SD (range) unless otherwise specified.

Abbreviations: BDT, Bangladeshi taka; SES, socioeconomic status.

* $P < .05$.

Description of the BGD MAL-ED Study

The MAL-ED study in Bangladesh is comprised of the following components: (1) birth cohort component, (2) case-control component, and (3) twin studies.

In the birth cohort component, 265 healthy newborn children living in Bauniabadh, Mirpur, were enrolled. Enrollment started in February 2010 and was completed in February 2012. Children were enrolled within the first week of birth, and visited every other day by MAL-ED field research assistants who interviewed the parent or caregiver.

To overcome the Hawthorne effect, a case-control study component was added. For the case-control study, cases were severe to moderately underweight children (weight-for-age z score [WAZ], < -2 based on the new World Health Organization growth reference standards [13]) aged 6–24 months. Controls were well-nourished children (WAZ > -1) matched for sex and area of residence. Five hundred case and 500 control children were initially planned for enrollment; however, 500 cases and only 480 controls were enrolled during the period of February 2010–February 2012.

In the twin cohort study, women with twin pregnancy were identified at Radda Maternal and Child Health–Family Planning Clinic, Mirpur, and enrolled. Identification of twin pregnancies was done by experienced physicians at the Radda Clinic on the basis of routine procedures of clinical assessment and abdominal ultrasonography. A total number of 27 pairs were screened, and 14 pairs were enrolled and followed up at regular intervals for 12 months for gut function, stool microbiological, nutritional, and clinical assessments.

Enrollment in the BGD MAL-ED Study

Before enrollment of children into the MAL-ED Bangladesh study, a census was carried out to identify children and pregnant women in the cohort community in Mirpur. All the children aged 6–24 months in the community were screened for WAZ.

Birth Cohort Study

MAL-ED study field-workers visited the cohort households within 17 days of delivery to screen mothers and children for possible entry into the study. On average, 10 children were recruited per month for the birth cohort component.

The MAL-ED study had a well-defined recruitment protocol with very stringent inclusion and exclusion criteria. For instance, mothers were asked before enrollment whether the family had plans to move outside the community. This included planned absence from the study area of >30 days, which would have made it difficult for the field-worker to contact the mother or caregiver during that time. Exclusion criteria for cohort recruitment were maternal age of <16 years, not a singleton pregnancy, another child already enrolled in the MAL-ED study, severe disease requiring hospitalization prior to recruitment, and severe acute or chronic conditions diagnosed by a physician (eg, neonatal disease, renal disease, chronic heart failure, liver disease, cystic fibrosis, congenital conditions).

Case-Control Study

Children were excluded from enrollment if they had any features suggestive of illnesses that might impact nutritional status or response to treatment of malnutrition (eg, severe diarrhea or pneumonia at the time of enrollment, persistent diarrhea, cleft lip or palate, blindness, tuberculosis, jaundice, renal or cardiac

disease, cerebral palsy, or chromosomal disorders including trisomy 21).

Treatment of Identified Malnutrition in the Case-Control BGD MAL-ED Study

In the case-control study component, cases received the standard nutritional supplement recommended by the National Nutrition Program of Bangladesh. The supplement, known as a *Pushti* packet, is a sachet containing 20 g roasted rice powder, 10 g roasted lentil powder, 3 g soybean oil, and 5 g molasses. The cases received the supplement as on-site feeding 6 days a week with severe underweight children (WAZ < -3) receiving 3 sachets (450 kcal), and moderate underweight children (WAZ -2 to -3) receiving 2 sachets (300 kcal). The provision of the supplement was continued for 5 months or until graduation when the child achieved WAZ -2 (or WAZ -1 for moderate malnutrition).

All children enrolled as cases and controls received counseling for mothers on improving child's nutritional status; high-potency vitamin A capsules at 6-month intervals (100 000 IU for children 6–12 months old and 200 000 IU for older children); deworming at enrollment; and a multiple micronutrient powder (MNP). Each MNP sachet contained 12.5 mg iron, 5 mg zinc, 300 μ g vitamin A, 150 μ g folic acid, and 50 mg of vitamin C. For the first 392 children enrolled, the MNP was provided for 2 months, whereas for all later recruitment, the MNP was provided for 4 months. Immunization with Expanded Program on Immunization vaccines was ensured for all enrolled children.

Control children did not receive any nutritional supplements (eg, *Pushti* packet or MNP sachet). Any child diagnosed to have severe acute malnutrition was admitted to the icddr,b hospital and treated according to the icddr,b standardized management protocol [14].

Quality Control Measures

The quality of observations of MAL-ED study is maintained by an in-built quality control and monitoring system involving all components of the study.

At the field level, data collection is monitored by different levels of supervisors. The activities of health workers are monitored and supervised by field research assistants; their immediate supervisors (field research supervisors) then re-collect 10% of all collected data including anthropometry and dietary assessments. Similarly, 10% of cognitive tests are re-performed by a senior psychologist. The activities of all field staff are supervised by 2 field research officers. They prepare weekly reports for the investigators and collaborate with laboratory and data management staff to maintain quality and synchronize activities.

Several forms addressing quality control issues have to be filled in for all biological samples collected from the field. To

ensure seamless data collection and in time we use a scheduler software to develop data and sample collection schedule for each child. All information, measurements and biological samples are collected as per the MAL-ED Manual of Procedures (MOP), and conform to icddr,b ethical review committee rules. The site principal investigator and 5 coinvestigators are responsible for overall site supervision and monitoring activities as per their respective professional competence. For example, there are cognitive, surveillance, data management, gut function, nutrition and vaccine response teams supervised by each coinvestigator. Moreover, 1 coinvestigator is responsible for coordination of all field, laboratory, and data management activities and serves as the site manager. The investigators also organize regular refresher training for all field staff and their supervisors. Finally, the site principal investigator oversees all activities of the study site.

At the highest level, The MAL-ED Administrative Core is responsible for general scientific oversight of the different locations, and evaluates progress through semiannual and annual reporting as well as site visits and meetings with site staffs. The Scientific Advisory Committee advises the Administrative Core to (1) provide overall guidance on the study protocols; (2) evaluate the progress of the project; and (3) aid in dissemination of findings as warranted. The Administrative Core monitors and evaluates all major activities on the basis of the project timeline and milestones. A common MOP containing standardized operating procedures and case report forms used in the study, as well as necessary training and quality assurance/quality control procedures to ensure comparability of results across the sites, was developed following extensive discussion between experts at the field sites and the technical subcommittee members. A protocol for real-time data transfer to and from each site to a centralized database was established to collate de-identified data for quality control review and cleaning.

Services to the Community

The BGD MAL-ED study provides access to healthcare services through its onsite clinic. The clinic is staffed with a physician 6 days per week who provides free care to the enrolled children and their immediate family members, with healthcare available at the icddr,b hospital during hours that the clinic is closed.

When diarrheal or respiratory disease is detected, the child is examined and treated as needed. As appropriate, a stool sample or nasopharyngeal swab is collected, kept on ice, and transported to the laboratory for processing within 2–4 hours. Parents are instructed in the use of oral rehydration salt solution. Appropriate antibiotics and other medications (ie, zinc) are dispensed free of cost when necessary. Children requiring hospitalization are also subsidized in seeking admission to a hospital or purchasing medicines.

Additional Value of the Case-Control Component

By using a multidisciplinary framework, the MAL-ED case-control study in Bangladesh is expected to provide insights into the genetic basis of malnutrition through genome-wide association scans, and into why some children fail to improve despite receiving the appropriate treatment for malnutrition. This study will identify the risk factors of malnutrition and its associated impact on child health employing conventional epidemiologic and novel genomic methods. By defining the risk factors and biomarkers for malnutrition, optimized strategies for interventions for childhood malnutrition, testing, evaluation, and surveillance can be developed [15].

Although only the cohort design can adequately assess sequential events that precede the onset of persistent diarrhea and/or malnutrition or growth shortfalls, close biweekly household follow-up is necessary for obtaining accurate data on diarrheal illnesses but may have a Hawthorne effect that dramatically reduces diarrhea rates and malnutrition, as well as mortality. The case-control study is expected to provide robust supplementary evidence to support the understanding of the etiology and pathogenesis of and interventions for moderate malnutrition.

CONCLUSIONS

Nearly one-third of children in low- and middle-income countries are malnourished. Malnutrition in the first 2 years of life leads to irreversible damage to cognitive function and physical capacity, and is transmitted between generations as malnourished mothers give birth to low-birth-weight children. The MAL-ED study seeks to gain a better understanding of the risk factors for malnutrition, enteric diseases, and associated health consequences, including developmental impairment, in children in low- and middle-income countries. The high prevalence of malnutrition and infectious morbidity, suboptimal sanitation, and high population density makes Bauniabadh in Mirpur, Bangladesh, an important site for contributing to the MAL-ED study.

Notes

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