

A Pilot School Meal Program Using Local Foods with Soybean in Rural Bangladesh: Effects on the Nutritional Status of Children

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Abstract

Background: Growth retardation is a challenge in Bangladesh. School feeding programs with fortified biscuits have been evaluated in Bangladesh. However, the impacts of a school meal program using local foods on the growth and nutritional status of children have not been investigated. **Objective:** To determine whether a school meal program (SMP) using local foods with soybean could improve children's growth and micronutrient status in rural Bangladesh. **Methods:** Two primary schools were randomly assigned as intervention (SMP; n = 200) and control (non-SMP; n = 200) schools. Children in the intervention school were supplied a school meal with local foods including soybean, containing more than one-third of the recommended daily allowance of energy and nutrients, 5 days/week for 8 months. The attendance rate and school lunch consumption of the children were monitored. Baseline and final anthropometry, hemoglobin and micronutrient status were assessed. **Results:** There were no significant differences in anthropometric measurements at baseline between the intervention and control groups, but there were differences in the prevalence of anemia, vitamin A deficiency and zinc deficiency. After the intervention, children in the SMP school showed a larger degree of improvement in the height-for-age Z-score ($P < 0.001$), red blood cell count ($P = 0.001$) and hemoglobin concentration ($P < 0.001$) than children in the non-SMP school. However, there were no positive effects on the body mass index (BMI)-for-age Z-score, serum ferritin, serum retinol or serum zinc status among children in the SMP school. **Conclusion:** A school meal program using local foods with soybean improved the height velocity and hemoglobin

concentration of children in rural Bangladesh.

Keywords

School Meal Program, Local Food, Child Growth, Anemia, Bangladesh

1. Introduction

Growth retardation, which includes stunting, wasting, underweight and micronutrient deficiency, continues to be a major public health problem in most low-income countries. Growth retardation and micronutrient deficiency contribute to mortality, morbidity, reduced immune competence and impaired cognitive function and productivity [1] [2] [3]. Growth retardation is generally caused by protein-energy malnutrition, and an estimated one-third of children under the age of 5 worldwide (178 million) are stunted, with 112 million underweight [4]. A short maternal stature increases the risk of small-for-gestational-age newborn and preterm birth [5]. The four most common forms of micronutrient malnutrition are iron, vitamin A, iodine and zinc [4]. Iron deficiency, the most common micronutrient deficiency in the world, causes anemia, which is a risk factor for maternal death and impaired physical and cognitive development and suboptimal immune systems in children [6]. Vitamin A deficiency, the second-most common micronutrient deficiency, is associated with an increased rate and severity of infectious diseases and is a primary cause of childhood morbidity and mortality, blindness in children and anemia [6] [7].

A number of trials of supplements for pregnant women and children under 5 years of age have been performed [8] [9] [10] [11]. However, fewer studies have been conducted in school-age children. Two main types of studies of the effect of school meals on the nutritional status of children have been performed. The first type includes studies of the effects of micronutrient fortification of school meals using seasoning powder. Such studies have been conducted in India [12] [13], Thailand [14] [15] [16], Indonesia [17], the Philippines [18], Himalayan villages [19] and Bangladesh [20]. Fortification interventions include trials of a micronutrient-fortified beverage in Tanzania [21] and Botswana [22], and micronutrient-fortified biscuits in South Africa [23], Vietnam [24] [25] and Haiti [26]. All of these studies suggested that such micronutrient fortifications improve micronutrient levels and short-term cognitive function. These approaches are suitable for high-risk areas/groups of school children but are not practical and not sustainable for all school children in developing countries.

The second type of study involves the effects of school meals. School meals made with local food are sustainable, and nutritionally appropriate school meals can be used to provide nutrition education to children and their families. They can even be an incentive for children to attend school and can activate the local food economy. However, few studies on the effects of school meals on the nutri-

tional status of children have been reported. In Jamaica, the provision of a school breakfast benefited children's school attendance, achievements and nutritional status, including height, weight and body mass index (BMI) [27] [28]. A community-based school snack was provided by the Indonesian government from 1996 to 1998. Its effects on the attendance rate of children, selection of nutritious foods by parents and community members, and benefit for the local economy via the use of local foods were reported, but no effects on nutritional status were reported [29]. A small-scale study in Indonesia found that school meals improved hemoglobin and hematocrit levels and body mass index [30], and a large-scale study in Pakistan found a decreased percentage of children with wasting [31], but no comparisons were made with controls. In Malawian children, school feeding was associated with an improvement in reversal learning and catch-up growth in lean muscle mass but not in height and weight [32]. Some studies used local foods, which contain nutrients showing shortages in children in low-income countries. In Burkina Faso, a randomized controlled intervention trial found a positive impact of the addition of red palm oil, a local food, to school meals on vitamin A status but not on anthropometry [33]. Many studies have been conducted in Kenya. An intake of micronutrients high in animal-derived foods was associated with better growth in rural Kenyan school children [34]. In addition, nutrient intake and nutritional status were better among participant pupils of a parent-supported school lunch program in Kenya compared with nonparticipant pupils [35]. Food supplements had a positive impact on weight gain, and the addition of animal-sourced foods increased the lean body mass of school children in Kenya [36], with meat supplementation also increasing arm muscle area [37]. A cluster randomized controlled feeding intervention showed beneficial effects of school snacks using meat, milk and vitamin A-fortified oil on morbidity [38] and school test scores [39].

A review of the effects of school meals indicated relatively consistent positive effects of school feeding on energy intake, micronutrient status, school enrollment and child attendance. However, their impact on growth, cognition and the academic achievements of school children is less clear [40] [41]. More evidence of the effects of school meals with local food on growth should be obtained.

Protein energy and micronutrient deficiency are serious public health problems in Bangladesh. The prevalence of stunting (height-for-age Z-score < -2), wasting (BMI-for-age Z-score < -2) and underweight (weight-for-age Z-score < -2) among children have been reported for under 5 years of age [42] and for 6 to 9 years of age [43]. Anemia is common among all age groups and in both sexes [44] [45] [46]. Subclinical vitamin A deficiency among preschool and school children has been reported [46] [47]. One of the causes of growth retardation and anemia is poor macro- and micronutrient intake. The daily intakes of energy per head for children aged 7 to 9 years and 10 to 12 years in 1995-1996 were 1504 kcal (males) and 1314 kcal (females) and 1788 kcal (males) and 1639 kcal (females), respectively [43]. The average protein intake in children aged 7 to 9

years in 1995-1996 was lower than required, at 37 g (males) and 33 g (females) [39]. Further data show that 20% - 40% of school children (aged 5 - 10 years) take less protein than required [48]. For adults, vitamin A deficiency is highly prevalent among rural pregnant women [49] [50]. The high prevalence of inadequate micronutrient intake has been shown by food balance sheets for Bangladesh [51]. The very poor micronutrient intake of preschool children and women is explained by both a low food intake and limited diversity [52].

Based on the previous data and higher primary school enrollment rate, at 93% for males and 97% for females in 2010-2014 [53], a school meal program with local foods would be a reasonable approach in Bangladesh. School feeding programs with fortified biscuits have been evaluated in Bangladesh [48] [54]. However, the impacts of a school meal program using local foods on the growth and nutritional status of children has not been investigated. We focused on protein, iron, zinc and vitamin A because these nutrients are particularly important for growth and anemia prevention, both of which are serious public health problems in Bangladesh, as mentioned above. To increase the amount and quality (amino acid variety) of protein and the amount of iron and zinc, we used soybean because the typical diet in Bangladesh is based on cereal, which can lead to inadequate protein consumption because of poor digestibility and limited amounts of lysine [55]. Many kinds of beans are used in Bangladesh, and soybean can easily be introduced into the local diet. Soybean is easy to grow, rich in lysine and can compensate for the shortage of lysine in rice. In addition, dried soybean can be preserved without refrigeration, which is important because many villages in Bangladesh lack electricity. To increase the amount of vitamin A, we also used local green leafy vegetables.

Thus, the objective of this study was to assess the impact of a school meal program with local foods containing soybean on the growth and micronutrient status of school children in rural Bangladesh.

2. Methods

2.1. Study Sites and Participants

The study was conducted in the Sharsha sub-district of the Jessore district, located in the southwestern region of Bangladesh bordered by India. A non-governmental organization, the Japan-Bangladesh Cultural Exchange Association (JBCEA), has been conducting an income-generating and life-style-improving program in this area. This study was a collaboration of the authors and the JBCEA.

Two government schools were selected according to the following criteria. We selected six areas in the Sharsha sub-district that were not beside the main road to avoid the bias of the introduction of new external food/information during the intervention. A sample size of 95 children per group was calculated based on an estimated difference in hemoglobin concentration at the end of the study of 4 g/L with an estimated standard deviation (SD) of 9 g/L, significance level

(two-tailed test) of 5% and power of 80%, with an additional 20% allowance for dropouts. One school with approximately 200 students (100 male and 100 female) was selected from each of the six areas. We measured all children's weight and height in the six schools and selected two of these schools without differences in the mean weight and height.

One school was randomly allocated as the intervention school, which was supplied the school meal (SMP school), and the other was the comparison school, which was not supplied the school meal (non-SMP school). The non-SMP school planned to introduce a school meal in the next year, and they agreed to join this study. There were 211 (98 males and 113 females) and 200 (111 males and 89 females) children registered in the SMP and non-SMP schools, respectively (**Figure 1**). All parents of the students were contacted and consented to their child's participation in the study. We excluded 13 children from each school who did not attend the baseline examination, as well as 2 children from the SMP school and 1 child from the non-SMP school who were not 5 to 13 years old. Thus, 193 children in the SMP school and 186 children in the non-SMP school were enrolled in the study. In addition, 20 children in the SMP school and 4 children in the non-SMP school did not attend the final examination. Accordingly, 172 children (78 males and 94 females) and 182 children (103 males and 79 females) completed the study in the SMP and non-SMP schools, respectively. The children who dropped out did not differ from the remaining children in any background characteristics or nutritional status. The schools have five classes corresponding with grades 1 through 5. The school year starts in January. The school management committee cooperated with this study.

2.2. Research Design

The baseline survey was conducted on February 17th and 18th, 2010. The school meal was served from March 1st, 2010 to November 8th, 2010. The end line survey was conducted on November 9th and 10th, 2010.

Children in the SMP school received the school meal at lunch time every day that they attended school. There were 253 days during the intervention period and 127 school days in the SMP school after excluding Fridays, Saturdays and holidays. There were 152 school days in the non-SMP school. The JBCEA staff checked the quality and quantity of the school meals at least once per week.

We created two kinds of meals based on the local menu that was popular in the village. One was "Kichuri", which is boiled rice porridge with vegetables, soybean and spices; the other was curry and boiled rice with soybean. The same ingredients were used in the two menus. The school meal ingredients are shown in **Table 1**. To make the school meal system sustainable, all foods were locally available at low cost. The average total cost including the cost of food, cooking fuel and payment for cooks per meal per child was 21.5 taka (this includes 15.4 taka for food), or approximately US \$0.3.

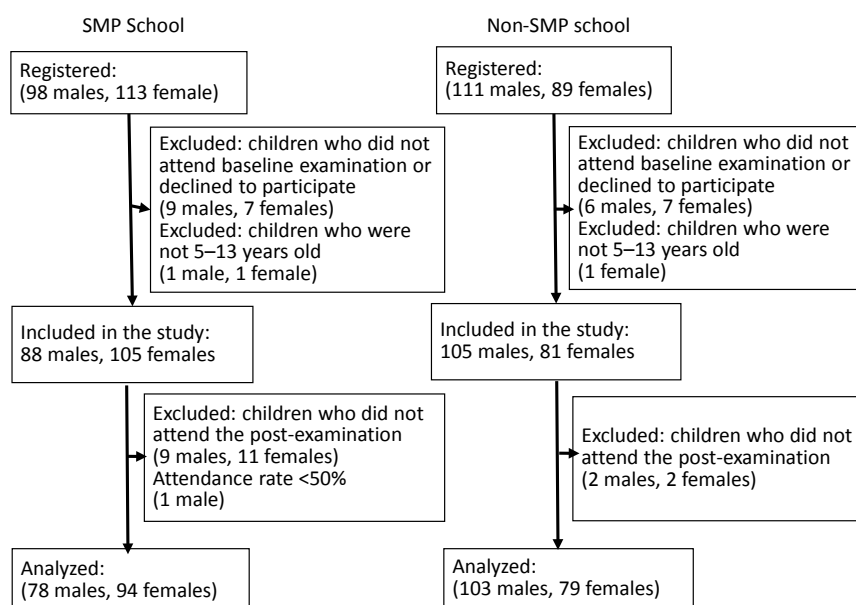


Figure 1. Trial profile.

Table 1. Amount of school meal ingredients per meal per child.

Food stuff	Class 1 and 2 ^a (g)	Class 3 - 5 ^b (g)
Rice	41.5	50
Soybean	34.5	42
Onion	16.6	20
Garlic	8.3	10
Chili	2.9	3.5
Ginger	3.3	4
Potato	41.5	50
Papaya	41.5	50
Pumpkin	83.5	100
Amaranthus tricolor (Joseph's coat)	49.8	60
Vegetable oil	11.6	14
Salt	3.5	4.2
Garam masala	0.4	1
Turmeric	0.7	1
Cumin	0.8	1

^aClass 1 and 2: 5 - 8 years old; ^bClass 3 - 5: 9 - 13 years old.

Before the school meal program was started, the nutrient values of the school meal and locally produced soybean were analyzed in Japan (Japan Food Research Laboratories) in August 2009. The energy and nutrient requirements for Bangladeshi children and the energy and nutrient contents of the school meal and soybean per person are shown in **Table 2**. The school meal supplied one-third of the daily energy and riboflavin requirements and more than two-thirds of the daily requirements for other nutrients based on the Recommended Dietary Allowances (RDAs) for Bangladeshi people [56]. Almost the same daily amounts of amino acids, based on a WHO report [55], were supplied

Table 2. Energy and nutrient requirements and composition of the school meal supplied at the SMP school.

Table Head		Classes 1 and 2 (5 - 8 years old)				Classes 3 - 5 (9 - 3 years old)			
		RDA /day ^a	Supplied by school meal ^b	Supplied by soybean ^c	% RDA ^d	RDA/day ^a	Supplied by school meal ^b	Supplied by soybean ^c	% RDA ^d
Energy	kcal	1820	631	136	35	2190	761	157	35
Protein	g	31.0	27.7	12.3	89	38.0	33.4	14.2	88
Fat	g	-	21.7	6.8	-	-	26.2	7.9	-
Carbohydrate	g	-	91.5	9.2	-	-	110.3	10.7	-
Sodium	mg	-	1850	0	-	-	2229	0	-
Potassium	mg	-	1702	535	-	-	2051	620	-
Calcium	mg	450	296	69	66	450	357	80	79
Iron	mg	10.0	9.0	1.9	90	10.0	10.0	2.2	100
Zinc	mg	-	2.6	1.2	-	-	3.2	1.4	-
Vitamin A	µg RE	300	328	0.0	109	400	395	0	99
Thiamine	mg	0.90	0.70	0.2	78	1.20	0.85	0.26	71
Riboflavin	mg	1.00	0.30	0.1	30	1.20	0.37	0.09	31
Folate	µg	100	152	0	152	100	183	0	183
Vitamin C	mg	20	33	0	165	20	40	0	200
Fiber	g	-	19.1	5.8	-	-	23.1	6.7	-
Amino acids									
Isoleucine	mg	961	1131	587	118	1178	1363	680	116
Leucine	mg	1891	1197	1011	63	2318	2406	1172	104
Lysine	mg	1488	1523	838	102	1824	1836	972	101
Sulfur amino acids	mg	744	846	380	114	912	1019	440	112
Methionine	mg	-	407	179	-	-	490	208	-
Cysteine	mg	-	439	200	-	-	530	232	-
Aromatic amino acids	mg	1271	2418	1321	190	1558	2913	1532	187
Phenylalanine	mg	-	1373	687	-	-	1654	796	-
Tyrosine	mg	-	1045	459	-	-	1259	532	-
Threonine	mg	775	1071	545	138	950	1291	632	136
Tryptophan	mg	205	373	176	182	251	449	204	179
Valine	mg	1240	1353	624	109	1520	1630	724	107
Histidine	mg	496	715	362	144	608	861	420	142
Arginine	mg	-	2081	980	-	-	2507	1136	-
Alanine	mg	-	1326	583	-	-	1598	676	-
Asparagine	mg	-	3371	1577	-	-	4061	1828	-
Glutamine	mg	-	5101	2498	-	-	6146	2896	-
Glycine	mg	-	1211	587	-	-	1460	680	-
Proline	mg	-	1339	697	-	-	1613	808	-
Serine	mg	-	1427	704	-	-	1719	816	-

^aRecommended dietary allowances (RDA) for Bangladesh children 4 - 6 years old and 7 - 9 years old (Institute of Nutrition & Food Science, University of Dhaka, 1992) were adopted for the RDAs of energy and nutrients for class 1 - 2 and class 3 - 5, respectively. - ; RDA is not available. Amino acids (mg) = Amino acid scoring pattern (mg/g protein requirement) for children 3 - 10 years old * protein requirement (g/day) for each age group. Protein and amino acid requirements were taken from Human Nutrition, a report from a joint WHO/FAO/UNU expert consultation, from the WHO technical report series 935, 2007. ^bEnergy and nutrients supplied by the school meal were analyzed in Japan Food Research Laboratories. ^cSoybean grown in the study village was analyzed in Japan Food Research Laboratories. Sulfur-containing amino acids = methionine and cysteine; Aromatic amino acids = phenylalanine and tyrosine. ^d% RDA = amount of nutrients supplied by the school meal/RDA * 100.

by the meal. The serving size of the school meal was different for classes 1 and 2 (5 - 8 years of age) and classes 3 through 5 (9 - 13 years of age) due to the differences in their RDAs. The nutrient contents of the school meal and % RDA are shown in **Table 2**.

Before the school meal program was started, we prepared the following: 1) a manual for school meal preparation, which included information on hygienic food preparation, cooking equipment, table wear, hand washing, order of foods, cooking methods and weighing and serving of meals; 2) training for chefs, the JBCEA employed and trained five chefs from the community for 5 days (3 days for hygiene and 2 days for cooking and serving), and four practice simulations involving meal cooking and serving were performed before the study began; and 3) cooking environment; as such, a kitchen and dining room were built at the school, and the water was tested to prevent arsenic contamination.

Regarding the time and place of the school meal, children in classes 1 and 2 had the meal from 12:15 to 13:15 in the classroom and went home afterward. Children in classes 3 through 5 had the meal from 11:00 to 12:00 in the dining room. The children in both schools took a tablet for deworming every 6 months. The SMP school children were instructed to wash their hands before eating the school meal.

The estimated school meal consumption was monitored according to the following methods. Teachers recorded the attendance of children every day during the intervention period. To estimate the consumption and calculate the consumption rate for each child, we weighed the serving amount and the leftovers for each child on 3 non-consecutive days in October 2010. The consumption rate was calculated by subtracting the leftover amount from the serving amount and dividing by the serving amount.

2.3. Data Collection

Sociodemographic data were collected at baseline only. Anthropometrical data and blood samples were collected from all children at baseline and at the end of the study.

Sociodemographic data were collected in January 2010 at the participants' homes. The staff of this project attended three training sessions of 2 hours each. They interviewed the parents of the children using a questionnaire. The questionnaire assessed household members, parents' education level, religion, household income, area of land owned by the household and number of months of rice shortage. To assess the protein intake levels, parents were asked the frequency of consumption of protein-rich foods (e.g., fish, egg, poultry and meat) of the child.

Anthropometric measurements (height and weight) were obtained by the same well-trained project staff from the JBCEA at baseline and at the end of the intervention period according to the anthropometric standardization reference manual [57]. All measurements were performed in the morning before lunch.

The height of the children was measured without shoes to the nearest 0.1 cm by a researcher using a Martin anthropometer. Height was measured twice, and if there was a difference > 0.5 mm, a third measurement was taken. Weight was measured to the nearest 0.05 kg on a digital scale (InnerScan SOV BC621, TANITA, Tokyo, Japan) with the children wearing light clothes. Anthropometric data were calculated as Z-scores for weight-for-age (WAZ), height-for-age (HAZ) and BMI (kg/m²)-for-age (BMIZ), based on the WHO Child Growth Standards for children aged 5 - 19 years [54]. The WAZ was calculated until 120 months of age because the standard was not available for children over 120 months of age.

Biochemical assessment was performed in the Bangladesh Institute of Research and Rehabilitation for Diabetes, Endocrine and Metabolic Disorders (BIRDEM). A non-fasting venous blood of approximately 7 - 8 mL was taken in the morning from each child by venipuncture using polypropylene syringes and divided into two tubes. The first tube (containing EDTA) was used for the estimation of hematological parameters including red blood cell (RBC) count, hemoglobin, hematocrit and mean corpuscular volume (MCV). The hematological tests were done on the same day in a certified diagnostic center of Jessore metropolitan city using a CELL-DYN 3200 System (Abbott Laboratories, Abbott Park, IL, USA). The remaining blood (6 mL) was then ejected into a glass tube for biochemical analysis. The blood sample was allowed to clot for approximately 30 min. The serum was separated from the coagulated blood in the field by a portable, battery-powered centrifuge at 3000 rpm for 30 min. The serum was aliquot into five different Eppendorf tubes with equal amounts of approximately 500 μ L in each tube; the vitamin A aliquot was collected in duplicate and was covered with aluminum foil due to light sensitivity. All samples were frozen on-site using dry ice and transported to BIRDEM, Dhaka, Bangladesh, where it was preserved at -56°C until analysis. Serum iron, total iron binding capacity (TIBC) and unsaturated iron binding capacity (UIBC) were measured using a bichromatic (600, 700 nm) endpoint technique. Serum ferritin was measured by a one-step enzyme immunoassay. Biochemical parameters were estimated by an automated analyzer (Dimension RxL Max, Siemens Healthcare Diagnostics Inc., USA). Serum retinol determinations were performed with a commercially available vitamin A and vitamin E determination kit (Bio-Rad A/E, Bio-Rad Inc., Hercules, CA, USA) by the use of the high-performance liquid chromatography (HPLC) method (Shimadzu, LC, Kyoto, Japan). Serum Zn was measured using a standard procedure involving flame atomic absorption spectrometry, which was modified from the graphite method (Perkin Elmer, AAnalyst 800, Shelton, CT, USA).

The precision of all of the biochemical assays was checked using pooled serum, and their accuracy was established using certified reference materials or appropriate manufacturer controls. The corresponding between-assay CVs of the low-to-high control samples for iron, ferritin, TIBC and UIBC were 4.2% to 1.8%, 7.8% to 6.5%, 2.8% to 3.97% and 2.1% to 2.9%, respectively. To estimate

serum zinc, zinc standards were prepared by diluting the stock standard solution [5% (v/v) glycerol]. A 5% (v/v) glycerol solution was used as a blank solution when determining serum zinc. The between-assay CVs of low and high standards were 14.0% and 12.5%, respectively. The vitamin A and E quality control material provided by Bio-Rad was used to monitor the accuracy of the laboratory testing procedures for the analytes. The mean value (SD, CV %) was 81.6 µg/dL (10.8, 13.1%) compared with the certified value of 78.0 (range 58 - 98) µg/dL.

2.4. Statistical Analysis

Stunted growth was defined as HAZ < -2 SDs and wasted was defined as BMIZ < -2 SDs of the WHO standards [58]. Anemia was defined as a hemoglobin concentration < 115 g/L for children aged 6 - 11 years and < 120 g/L for children aged 12 years and older [59]. A low MCV was defined as an MCV < 80 fL, and plasma ferritin concentrations < 20 µg/L were defined as depleted [59]. Subclinical vitamin A deficiency was defined as a plasma retinol concentration < 0.70 µmol/L [60]. Zinc deficiency was defined as serum Zn < 0.65 mg/L for children < 10 years, Zn < 0.66 for female subjects ≥ 10 years and Zn < 0.70 mg/L for male subjects ≥ 10 years [61].

Statistical analyses were performed using SPSS software (Windows, version 21.0). Differences in sociodemographic characteristics and the prevalence of undernutrition at baseline between children in the SMP and non-SMP schools were tested using chi-square and t-tests (two-tailed). Differences in baseline data between children in the SMP and non-SMP schools were examined using t-tests (two-tailed). Differences in changes in the data from baseline to endline between children in the SMP and non-SMP schools were examined by ANCOVA, adjusted for sex, age and value at baseline. Differences were considered significant at $P < 0.05$.

The study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects were approved by the ethics committee of the Niigata University of Health and Welfare, Japan (No. 17155-100106), and the National Research Ethics Committee of Bangladesh in Medical Research Council (BMRC/NREC/2007-2010/147). The staff of this study visited all students' houses to explain the school meal and study protocol to the parents. Parents were invited to an information meeting at the schools. Written informed consent was obtained from all subjects' parents. Permission from the Jessore District, Sharsha sub-district, communities and schools were obtained via meetings. This trial was also approved by the Government of the People's Republic of Bangladesh, Office of the Primary Education Section 2 (memo no. PraShia/Pori: and Dev:/25/2010/62).

3. Results

3.1. Characteristics of Participants at Baseline and Attendance Rate/Consumption Rate during the Program

The sociodemographic characteristics at baseline are shown in **Table 3**. There

were no significant differences in sociodemographic status between the SMP and non-SMP school-children except for the ratio of males, area of household land and frequency of fish consumption. The ratio of male children was lower in the SMP school (45.3%) than in the non-SMP school (56.6%). The mean (standard error) ages of children in the SMP and non-SMP schools were 106.5 (1.8) months and 108.3 (1.8) months, respectively. The SMP school households owned a smaller area of land than non-SMP school households. Most of the children did not eat fish, egg, poultry or meat every day. The frequency of fish consumption was higher in children in the SMP school than in the non-SMP school.

The nutritional status of the children at baseline is shown in **Table 4**. Stunted growth was seen in 19.2% and 23.3% of males and 28.7% and 32.9% of females in the SMP and non-SMP schools, respectively. Wasting was found in 23.1% and 23.3% of males and 22.3% and 19.0% of females in the SMP and non-SMP schools, respectively. There were no significant differences between the SMP and non-SMP schools in terms of stunting and wasting prevalence at baseline. At baseline, anemia prevalence was significantly higher in the SMP school compared to non-SMP school, 83.3% vs 70.9% ($p < 0.001$), for males and 95.7% vs 81.0% ($p < 0.01$) for females, respectively. The prevalence of depleted iron stores was very low. Males in the SMP school showed a significantly lower prevalence of subclinical vitamin A deficiency and a higher prevalence of zinc deficiency than males in the non-SMP school.

The median (25th, 75th percentile) attendance rates at the SMP and non-SMP schools were 81.1% (74.8%, 87.2%) and 96.7% (93.4%, 98.0%), respectively. The median (25th, 75th percentile) consumption rate of the school meal at the SMP school was 78.6% (59.0%, 100.0%), and we estimated that the consumption rate was approximately 80%.

3.2. Effects of the School Meal on Anthropometric Outcomes

The anthropometric outcomes are shown in **Table 5**. There were no significant differences in anthropometric indicators at baseline between the SMP and non-SMP schools. After 8 months, children in the SMP school showed greater increases in height and HAZ than children in the non-SMP school, both males and females. The mean (standard error) changes in height and HAZ of children in SMP vs non-SMP schools were 3.8 (0.1) cm vs 3.3 (0.1) cm ($P < 0.001$) and 0.03 (0.01) vs -0.04 (0.01) ($P < 0.001$), respectively. There were no differences in mean changes in weight, BMI, WAZ or BMIZ.

3.3. Effects of the School Meal on Biochemistry Outcomes

The biochemical outcomes are shown in **Table 6**. There were significant differences in baseline data between the SMP and non-SMP schools for hemoglobin, UIBC, serum retinol and serum zinc. Children in the SMP school had lower levels of hemoglobin and serum zinc and higher levels of UIBC and serum retinol

Table 3. Baseline sociodemographic characteristics of the participants.

Variables		SMP (n = 172)	Non-SMP (n = 182)	<i>P</i> -value
Sex (%)	Male	45.3	56.6	0.043
Age at baseline (months)	mean (SE)	106.5 (1.8)	108.3 (1.8)	0.48
Number of family members (%)	3 - 4 persons	45.3	41.4	0.76
	5 - 6 persons	41.2	43.6	
	7 - persons	13.5	14.9	
Number of children at home (%)	1 - 2 children	52.4	54.7	0.74
	3 - 4 children	41.8	38.1	
	5 - children	5.9	7.2	
Birth order of the subject child (%)	1st child	41.2	39.8	0.29
	2nd child	28.2	35.4	
	3rd child	30.6	24.9	
Education level of father (%)	No education	46.5	44.8	0.46
	Primary school	31.2	38.1	
	Secondary school	12.9	10.5	
	High school and higher	9.4	6.6	
Education level of mother (%)	No education	31.2	32.0	0.72
	Primary school	44.1	39.2	
	Secondary school	18.8	23.2	
	High school and higher	5.9	5.5	
Household income per month (%)	Low income (0 - 2500 taka)	34.1	35.9	0.47
	Middle income (2501 - 3000 taka)	32.4	36.5	
	High income (3001 - taka)	33.5	27.6	
Own land (%)	0 - 14 katha	47.1	33.1	0.017
	15 - 43 katha	21.2	31.5	
	45 - 600 katha	31.8	35.4	
Number of months of rice shortage during the last year (%)	0 - 1 month/year	34.1	35.9	0.82
	2 - 5 months/year	21.2	22.7	
	6 months/year	44.7	41.4	
Frequency of fish consumption for the child (%)	Every day	23.8	13.5	0.032
	3 - 6/week	21.4	20.2	
	1 - 2/week	38.1	51.7	
	<1/week	16.7	14.6	
Frequency of egg consumption for the child (%)	Every day	2.4	1.1	0.73
	3 - 6/week	6.5	5.5	
	1 - 2/week	51.2	55.2	
	<1/week	40.0	38.1	
Frequency of poultry or meat consumption for the child (%)	Every day	0.6	0.0	0.51
	3 - 6/week	0.0	0.6	
	1 - 2/week	8.2	9.9	
	<1/week	91.2	89.5	
Frequency of diarrhea (%)	More than once/month	2.4	1.7	0.72
Frequency of fever (%)	More than once/month	8.2	5.6	0.59

SE: standard error, 1 taka = 0.01537 USD, 1 katha = 720 square feet (67 m²). ^aNumber of respondents to the questionnaire. ^b*p*-value for chi-square test between children in the SMP and non-SMP schools except age at baseline. *p*-value for Mann-Whitney U-test between children in the SMP and non-SMP schools for age at baseline.

Table 4. Prevalence of undernutrition at baseline.

Variables	Male			Female		
	SMP	Non-SMP	<i>p</i> -value ^a	SMP	Non-SMP	<i>p</i> -value ^a
	(n = 78) %	(n = 103) %		(n = 94) %	(n = 79) %	
Stunted (%)	19.2	23.3	1.00	28.7	32.9	0.25
Wasted (%)	23.1	23.3	0.22	22.3	19.0	0.29
Anemia (%)	83.3	70.9	<0.001	95.7	81.0	0.006
Depleted iron stores (%)	0.0	1.9	0.51	1.1	6.3	0.09
Subclinical vitamin A deficiency (%)	3.8	16.5	0.008	13.8	19.0	0.41
Zinc deficiency (%)	35.9	13.6	0.001	26.6	19.0	0.28

^a*p*-value for chi-square test between children in the SMP and non-SMP schools. Stunted was defined as a height-for-age Z-score < -2 SDs of the WHO standards median. Wasted was defined as a BMI-for-age Z score < -2 SDs of the WHO standards median. Anemia was defined as hemoglobin (Hb) < 115 g/L for boys and girls < 12 years old and Hb < 120 g/L for boys and girls ≥ 12 years old. Depleted iron stores were defined as serum ferritin < 15 µg/L in boys and girls of all ages. Subclinical vitamin A deficiency was defined as serum retinol < 0.7 µmol/L (20 µg/dL) in boys and girls of all ages. Zinc deficiency was defined as serum Zn < 0.65 mg/L for children < 10 years old, < 0.66 mg/L for girls ≥ 10 years old, and < 0.70 mg/L for boys ≥ 10 years old.

Table 5. Anthropometric status of participants at baseline and endline and the change between baseline and endline.

Variables	Time	SMP (n = 172)		Non-SMP (n = 182)		<i>p</i> -value ^a	<i>p</i> -value ^b
		mean	SE	mean	SE		
Height (cm)	Baseline	124.3	0.7	124.0	0.8	0.76	
	Endline	128.1	0.8	127.2	0.8		
	Change	3.8	0.1	3.3	0.1	<0.001	<0.001
Weight (kg)	Baseline	22.5	0.4	22.3	0.4	0.76	
	Endline	24.1	0.4	23.8	0.4		
	Change	1.7	0.1	1.5	0.1	0.07	0.15
BMI (kg/m ²)	Baseline	14.3	0.1	14.3	0.1	0.90	
	Endline	14.5	0.1	14.5	0.1		
	Change	0.2	0.0	0.2	0.0	0.94	0.74
Height-for-age Z-score	Baseline	-1.2	0.08	-1.40	0.08	0.07	
	Endline	-1.2	0.08	-1.50	0.08		
	Change	0.03	0.01	-0.04	0.01	<0.001	<0.001
Weight-for-age Z-score ^c	Baseline	-1.30	0.09	-1.50	0.08	0.12	
	Endline	-1.20	0.08	-1.40	0.08		
	Change	0.12	0.03	0.11	0.03	0.81	0.11
BMI-for-age Z-score	Baseline	-1.30	0.07	-1.30	0.06	0.68	
	Endline	-1.30	0.07	-1.30	0.06		
	Change	0.01	0.03	-0.01	0.02	0.74	0.92

SE: standard error. ^a*p*-value for t-test between children in the SMP and non-SMP schools. ^b*p*-value for ANCOVA adjusted for sex, age and value at baseline between children in the SMP and non-SMP schools. ^cThe numbers of participants in the SMP and non-SMP schools were 159 and 157, respectively, because WHO reference data is not available for children younger than 10 years old. Z-scores were based on WHO Child Growth Standards for children 5 - 19 years old (2007).

Table 6. Micronutrient status of participants at baseline and endline and the change between baseline and endline.

Variables	Time	SMP (n = 172)		Non-SMP (n = 182)		<i>p</i> -value ^a	<i>p</i> -value ^b
		mean	SE	mean	SE		
Hemoglobin (g/L)	Baseline	10.7	0.0	11.0	0.1	<0.001	
	Endline	10.6	0.1	10.5	0.1		
	Change	0.0	0.1	-0.5	0.1	<0.001	<0.001
Hematocrit (%)	Baseline	32.5	0.2	32.1	0.4	0.38	
	Endline	34.7	0.2	34.6	0.3		
	Change	2.3	0.2	2.6	0.4	0.51	0.80
Serum ferritin (µg/L)	Baseline	60.5	2.5	55.2	1.9	0.09	
	Endline	69.3	2.7	64.7	1.9		
	Change	8.8	2.9	9.4	1.9	0.86	0.45
Serum iron (µmol/L)	Baseline	11.1	0.3	11.8	0.3	0.10	
	Endline	15.2	0.3	14.9	0.3		
	Change	4.1	0.3	3.1	0.4	0.048	0.21
Red blood cell (RBC) (million/µL)	Baseline	4.0	0.0	3.9	0.0	0.23	
	Endline	4.1	0.0	4.0	0.0		
	Change	0.2	0.0	0.1	0.0	0.042	0.001
Mean cell volume (MCV) (fL)	Baseline	82.3	0.4	81.4	0.4	0.10	
	Endline	84.8	0.5	86.5	0.4		
	Change	2.5	0.3	5.1	0.3	<0.001	<0.001
Unsaturated iron binding capacity (UIBC) (µmol/L)	Baseline	54.5	0.9	52.2	0.7	0.041	
	Endline	50.5	0.9	47.9	0.8		
	Change	-4.0	1.3	-4.3	0.9	0.85	0.02
Total iron-binding capacity (TIBC) (µmol/L)	Baseline	65.0	1.0	63.6	0.6	0.22	
	Endline	63.3	0.7	63.1	0.7		
	Change	-1.7	1.1	-0.5	0.8	0.39	0.95
Transferrin saturation (%)	Baseline	17.9	0.7	18.8	0.4	0.22	
	Endline	24.5	0.6	24.1	0.6		
	Change	6.7	0.7	5.2	0.6	0.13	0.34
Serum retinol (µg/dL)	Baseline	37.6	1.3	31.8	1.0	0.001	
	Endline	41.6	1.5	36.3	0.7		
	Change	4.0	1.7	4.5	1.2	0.83	0.02
Serum zinc (mg/L)	Baseline	0.9	0.03	1.1	0.04	<0.001	
	Endline	1.2	0.04	1.4	0.04		
	Change	0.3	0.03	0.3	0.04	0.30	0.69

SE: standard error. ^a*P*-value for t-test between children in the SMP and non-SMP schools. ^b*P*-value for ANCOVA adjusted for sex, age and value at baseline between children in the SMP and non-SMP schools.

than children in the non-SMP school. After 8 months, children in the SMP school showed significantly larger amount of changes of hemoglobin and RBC levels than children in the non-SMP school. The mean (standard error) changes in hemoglobin and RBC levels of children in SMP vs non-SMP schools were 0.0 (0.1) g/L vs -0.5 (0.1) g/L (*P* < 0.001) and 0.2 (0.0) vs 0.1 (0.0) (*P* = 0.001), re-

spectively. In contrast, the changes in MCV ($P < 0.001$), UIBC ($P = 0.02$) and serum retinol ($P = 0.02$) were smaller in the children in the SMP school than in the children in the non-SMP school. There were no differences in the amount of change between the two schools for other indicators, such as hematocrit, serum ferritin, serum iron, TIBC, transferrin saturation and serum zinc.

There were no significant changes in the prevalence of stunting, wasting, anemia, depleted iron stores, subclinical vitamin A deficiency or zinc deficiency (data not shown).

4. Discussion

This study investigated whether the administration of a community-based school meal containing local foods and soybean over a period of 8 months could improve the physical growth and micronutrient status of school children. To our knowledge, this is the first study to use soybean and local foods without any fortification in a school meal program and to evaluate their impact compared with a control group. Children who participated in the school meal program showed significant improvements in height velocity, HAZ and hemoglobin concentration compared with nonparticipating children. These findings stress the importance of an adequate protein and amino acid composition of school meals in low-income countries where there is a low availability of animal protein foods.

Energy and nutrient intakes from the school meal in this study were estimated. The school meal supplied about one-third (33%) of the energy and riboflavin needs, more than two-thirds (66%) of other nutrients and almost the correct amounts of amino acids according to the daily requirements. Children consumed 80% of the supplied school meal per day. Among a total of 253 days during the intervention period, there were 127 school days, meaning that the school meal was only supplied on 50% of days. The estimated average requirement intake per day during the intervention period was 13% of energy and riboflavin, 26% of other nutrients and 40% of amino acids. Thus, we need to consider the number of school days when planning an effective school meal program.

At baseline, the rates of stunting and wasting were 20% - 30% and 20%, respectively. In previous studies, the prevalence of stunting in 2011 and 2014 was 41% and 16%, the prevalence of wasting in the same years was 36% and 14%, respectively, among children under 5 years of age [42]. In 1995-1996, the prevalence of stunting and wasting among children aged 6 to 9 years was 54.8% (males) and 50.1% (females) and 17.7% (males) and 19.1% (females), respectively [43]. Another study reported that the rates of stunting and wasting were approximately 20% and 30%, respectively, in 2008 [20]. The baseline anthropometric status of this study was thus similar to that of previous work.

The soybean-based school meal improved the linear growth of children with a statistically significant change in height velocity and HAZ, but not in BMIZ. The results are comparable to those of a previous one-year intervention study in Bangladesh school children using micronutrient-fortified yogurt [20]. However, there is little evidence on the effects of school meals on anthropometry out-

comes. Studies from Jamaica [27] and Malawi [32] showed no effects of school meals on weight and height, whereas a study from Indonesia and Pakistan showed an effect on recovery from wasting [30] [31]. Height gain during the school feeding intervention period, using milk or meat, was positively predicted by the average daily intake of energy from animal foods, iron, calcium, vitamin A and vitamin B₁₂ among Kenyan school children [34]. However, a review reported that the positive impact of school feeding on growth was less conclusive [40].

There are some possible reasons for these results. One reason is the effects of micronutrients. A systematic review noted that the effects of micronutrients on growth were equivocal [62]. A meta-analysis of the effects of micronutrients on the growth of children found that multi-micronutrient interventions did improve child growth [63]. Another possible reason is protein quality and quantity. Additional dietary protein, mainly from soybean, increases tissue deposition for growth and provides an important anabolic drive for linear bone growth in this study. Several intervention studies with additional protein-rich foods suggested a similar specific stimulatory effect of protein on linear bone growth mediated through IGF-1 [55]. In the current study, serum retinol and serum zinc levels were not improved in the children in the SMP school. Thus, it is reasonable to assume that the height gain during the school feeding intervention period using soybean was positively predicted by protein quantity and quality. Further research into the mediator between protein nutritional status and growth, such as IGF-1 and growth hormone, is needed to confirm this assumption.

The prevalence of anemia at baseline was 80% - 95% in the SMP school and 70% - 80% in the non-SMP school, showing a significant difference. In previous studies, the prevalence of anemia in preschool children, school children and pregnant women was 50% - 60%, 30% - 40% and 40% - 50%, respectively, from 2001 to 2004 [44] [45]. The prevalence of anemia in school children in 2008 was 53%, 16 and in 2011-2012, it was 19% [46]. The anemia prevalence in this study was thus higher than that of previous studies. However, the prevalence of depleted iron stores was very low. A high prevalence of anemia without depleted iron stores was observed in previous studies [20] [64] [65]. A high level of iron in groundwater could be one of the reasons for the higher iron status [64] [65]. The low hemoglobin concentration is explained by the low intake of animal foods and vitamin A [66] [67], and multiple micronutrient fortifications of food or supplementation enhanced the hemoglobin status in Bangladesh [68] [69]. Children in the non-SMP school showed a hemoglobin concentration decrease of -0.5 g/L during the intervention period, but the hemoglobin concentration of the SMP school children did not change; the difference in the changes between the groups was significant. One of the possible reasons for the decrease in the hemoglobin concentration among non-SMP school children might be an increased need for hemoglobin mass during rapid growth [60]. The SMP school children showed no change in hemoglobin concentration because the school meal compensated for the increased need for nutrients to increase hemoglobin

mass according to growth. The significant differences in the hemoglobin concentration change and RBC count between SMP and non-SMP school children, without a difference in serum iron status, suggests better utilization of iron, which may have been due to the presence of multiple micronutrients in the soybean school meal, as suggested by previous intervention studies [13] [14] [19] [20] [21] [23] [24] [25] [68] [69]. Another possible reason is the increased quality and quantity of the protein from the soybean school meal in the SMP school children. Protein quality and quantity, especially histidine, are important for maintaining the hemoglobin concentration [70].

The prevalence of subclinical vitamin A deficiency at baseline was less than 20%. The prevalence of zinc deficiency ranged from 13% to 36%. In previous studies, the rates of subclinical vitamin A deficiency among preschool children and school children were 56% and 3% - 4%, respectively, in the 1990s [47], and both were 21% in 2011-2012 [46]. The baseline nutritional status of vitamin A in this study was similar to that of previous studies. There was no previous data for zinc deficiency among children in Bangladesh. There were no positive effects of the school meal on serum retinol or zinc concentrations, possibly due to the relatively higher serum retinol and zinc concentrations at baseline and an insufficient amount in the meal to cause significant changes during the intervention period.

There are some limitations in this study. First, the baseline data of nutritional biomarkers in blood differed between intervention and non-intervention children because we ethically could not take blood before sampling the schools. Second, infection can influence biomedical indicators, but we did not measure C-reactive protein levels or monitor morbidity. Third, this study was conducted only for one intervention school and one non-intervention school in one area of rural Bangladesh.

Even with these limitations, the findings of this study suggest that school meals containing a certain amount of amino acids improve height velocity in school children in low-income countries. Not only iron but also protein may play important roles in maintaining hemoglobin concentrations. Thus, both micronutrients and the amount and quality of protein should be considered when providing school meals in low-income countries.

Two directions of further study are recommended. One is a large-scale, population-based study. Another is a study to investigate the mechanisms of the effects of a school meal with soybean on growth and the quality and quantity of protein required for appropriate human growth.

5. Conclusion

Children who participated in a community-based school meal program using local foods with soybean showed significant improvements in height velocity and hemoglobin concentration compared with children who did not participate in the school meal program. These findings support the positive effects of school meal program with local foods which contain adequate nutrients on improving

nutritional status of children in low-income countries where high prevalence of undernutrition.

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