

Effects of Pelagic Small Fish Powder on Nutritional Outcomes Among Children Aged 6-24 Months: A Quasi-Experimental Trial

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Abstract

Background

The first 1000 days of a child's life are a unique period of opportunity to lay the foundations for optimum health, growth, and cognitive development. After exclusive breastfeeding, complementary feeding is required to meet nutritional needs. This study aimed to observe the efficacy of a ready-to-use fish product (RUFPP) on nutritional outcomes of children aged 6 to 24 months in two communities in southeast coastal Bangladesh.

Methods

An experimental design was employed in which an intervention group (n = 30) received RUFPP in a proportionate combination of 5g anchovy (olua, *Coilia dussumieri*), sardine (chapila, *Sardinella longiceps*), and thryssa (faissa, *Thryssa dussumieri*) (40:30:30) for four months, while the control group (n = 27) received usual care. Anthropometric measures and biochemical markers (retinol, vitamin B12, ferritin, and iron), as well as secondary outcomes (hemoglobin, calcium, and random blood sugar (RBS)), were considered to assess differences between baseline and the 16-week post-intervention assessment.

Results

Mean (SD) age at baseline was 13.8 (4.4) months in the control group and 12.1 (3.7 months in the intervention group. Retinol levels in the intervention group increased significantly (25.9 ± 3.5 to 35.2 ± 2.2 µg/dl), surpassing the increase in the control group ($p < 0.05$). In the intervention group, the mean change in ferritin levels increased by 15.5 (18.4) ng/mL from baseline to post-intervention, which was significantly larger than the mean change in the control group (-1.3 (6.3) ng/mL) ($p < 0.05$). The intervention group showed a significant increase in hemoglobin levels ($p < 0.05$), whereas the control group showed a decrease. Besides, RBS were significantly decreased in the intervention group ($p < 0.05$), although between-group changes were not statistically significant ($p > 0.05$). However, no significant changes have been observed in serum iron, vitamin B12, or calcium status (except in the control group).

Conclusions

These findings suggest that the RUFPP may enhance the nutritional quality of complementary foods for children, thereby contributing to improved child health outcomes. However, given the limited sample size, these results should be interpreted with caution, and further studies with larger populations are warranted to confirm the observed effects.

Trial registration:

This trial is registered in the ISRCTN Registry (Registration ID: ISRCTN34731061, registration date 14.02.2025).

Introduction

The first 1000 days of human life, from conception to the second birthday of the child, often known as the 'critical window', are vital for children's growth and development (1). From a nutritional standpoint, two significant biological phases occur during the first 1000 days of an individual's existence. The first transition is the shift from intra-uterine nourishment to breastfeeding immediately after delivery. The second significant milestone is the introduction of

complementary food alongside breast milk throughout the weaning phase. While breast milk is widely acknowledged as the optimal source of nourishment for infants, it is insufficient to meet their nutrient requirements beyond the first six months of life. Hence, introducing complementary foods is customary for newborns in conjunction with breastfeeding, commencing at six months of age, to address nutritional deficiencies. Weaning food, despite its increased susceptibility, appears to receive considerably less attention, even though a substantial amount of research has been devoted to the significance of breast milk in promoting and supporting child development (2–4).

To guarantee adequate supplementary feeding for the optimal growth, maturation, and well-being of infants and young children, the WHO/UNICEF Global Strategy for Infant and Young Child Feeding (IYCF) encouraged the use of locally accessible foodstuffs (5) up to the age of two (6, 7). Children who consume foods with animal-source proteins in their diets are more likely to attain greater height than children who consume foods with plant-sourced proteins (8). Small pelagic fish, like anchovies and sardines, enhance the absorption of iron and zinc from plant-source foods (9). This finding is of great importance for populations facing micronutrient deficiencies, especially those who depend heavily on plant-based diets. Although animal-based foods like meat, poultry, and dairy products are beneficial for early complementary feeding, their high cost is a challenge for low-income households, hindering them from incorporating these nutritious options into their diet (10). According to the Bangladesh Demographic and Health Survey (BDHS) 2022, 61% of children aged 6–23 months were fed at least 3 times in line with the recommended IYCF practices, and 39% received an appropriately diversified diet. According to IYCF guidelines (11) 29% of infants and children aged 6–23 months received a minimum acceptable diet. Moreover, just 40% of children are given a diet that meets basic standards, even among families in the top quintile of income (12).

Using readily available marine fish, i.e., ready-to-use fish products (RUFs), rich in nutrients, may help overcome the difficulty of accessing expensive foods (13). Besides, inexpensive protein and micronutrient sources, such as fish, can be used in place of more expensive livestock diets (14). Fish and other aquatic foods are commonly available and meet the food preferences of children and adults in Bangladesh and most other developing countries. Fish is the foremost source of animal protein in coastal areas and is a nutrient-dense food group with high-quality protein (15). Studies from Bangladesh and Sri Lanka showed that fish, particularly small pelagic fish consumed whole, are good sources of protein and a range of micronutrients, including calcium, iron, zinc, iodine, vitamin A, vitamin D, and vitamin B₁₂. In addition, fish is a unique source of the long-chain n-3 polyunsaturated fatty acids (n-3 PUFA) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (16–18). Modeled evidence suggests that consuming mola (*Amblypharyngodon mola*), a common small fish in Bangladesh, may provide greater benefits and cost-effectiveness than a national vitamin A flour fortification program, as measured by disability-adjusted life years (19). Furthermore, introducing small freshwater fish or RUFs into the diets of children aged 6 months or older has been shown to enhance growth outcomes (20, 21).

RUFs are highly accepted by children (22, 23) showing a comparable impact to industrially processed supplements, with a limited effect on reducing growth faltering (22, 24). Promoting fish-based complementary foods is vital given the availability, affordability, and accessibility of various fish species in coastal locations, and their intake can be used as a successful food-based approach to improve the nutritional security of the local population (25). In this regard, RUFs can be added like spices to many types of cuisine, and their nutritional value and versatility may help them gain acceptance and popularity worldwide. These products may play a significant role in meeting a child's daily nutrient requirements. For example, RUFs can provide approximately 62% and 43% of the recommended daily allowance (RDA) of calcium and selenium, respectively, for adults; 83% and 58% for infants; and 71% and 45% for pregnant and lactating women (PLW) (13). In such a way, the significance of fish for food security and nutrition is becoming widely recognized as a potential source for alleviating hidden hunger (micronutrient deficiencies) (26).

This quasi-experimental study aimed to evaluate the nutritional effects of a newly developed RUFP among children under two years of age in Cox's Bazar, Bangladesh. The intervention group received the RUFP as a nutritional supplement. In addition, the study evaluated caregivers' acceptance of the RUFP for their children and their preferences regarding product marketing.

Methods

Study design and ethical consideration

A two-armed quasi-experimental study was conducted during October 2021 and January 2022. The intervention group ($n = 30$) received 5g of RUFPs made from anchovy (olua, *Coilia dussumieri*), sardines (chapila, *Sardinella longiceps*), and thryssa (faisaa, *Thryssa dussumieri*) daily in their complementary food at a 40:30:30 ratio, for 16 weeks. The control group ($n = 30$) was asked to maintain regular complementary food. Guided interviews were conducted pre- and post-intervention to obtain data on socio-economic factors, dietary intake, and anthropometric and biochemical assessments.

The principles outlined in the Declaration of Helsinki (27) served as the basis for the research. This trial is registered in the ISRCTN Registry (ISRCTN34731061, registration date 14.02.2025). Additionally, the Noakhali Science and Technology University Ethical Committee (NSTUEC) approved the research project (Reg: NSTU/SCJ/EC/2021/73) after receiving written consent from each participant. All participants were informed that they could withdraw from the study at any time without giving a reason. Informed parental consent was acquired from the enrolled children. This study followed a quasi-experimental design and is reported in accordance with the TREND (Transparent Reporting of Evaluations with Nonrandomized Designs) statement.

Sample size, study area selection, and enrollment

The study was conducted in Ukhiya and Teknaf sub-districts in Cox's Bazar district, Bangladesh. A total of 79 children were assessed for eligibility in this study, of whom 19 were excluded for not meeting the inclusion criteria ($n = 13$) or for unwillingness to participate ($n = 6$). The remaining 60 children were allocated to the study groups, as illustrated in Fig. 1. The control group comprised 30 participants recruited from the North Shilkhali Union of Teknaf sub-district, while the intervention group comprised 30 participants recruited from the Nidania Union of Ukhiya sub-district. Hence, 60 participants were recruited for this study. Selection of both sites was based on household-level survey data collected by WorldFish before the intervention, indicating that both intervened- and control households were similar in sociodemographic indicators (Table 1).

Village health volunteers (VHV) with expertise in community-based nutrition programs supported the project and engaged potential caregivers to allow their children to participate in the research. A data enumerator team visited the selected households and screened for the eligibility criteria, including child age, health condition, and willingness to consume RUFPs. Participants were instructed to maintain their normal lifestyle habits, including eating and playing regularly. were all healthy children aged 6–18 months. The exclusion criteria for enrollment of unwell children in the study were: unwell children, severely malnourished ($WHZ < -3$), overnourished ($WHZ > 3$), or seriously anemic ($Hb < 70$ g/L), based on a clinical screening process (24). Additionally, children with known dietary intolerance and/or enrolled in any form of supplementary feeding program were excluded from the study.

Nutrient composition of RUFPs and their contribution to the daily recommended allowance

After purchasing raw fish, it was transported and stored at 4°C in the analytical laboratory of the Department of Fisheries and Marine Science at Noakhali Science and Technology University, where it was cleaned and processed according to standard operating protocol to make RUFPs. Hygiene was maintained according to the instructions for a similar product (13, 28).

The macro- and micronutrients in the RUFP were analyzed using validated and standardized procedures. To ensure the reliability of the analysis, certified reference materials, recovery tests, and method detection/quantification limits were used.

The Association of Official Analytical Chemists (AOAC) International methods were used for analyzing protein composition (29). These proximate composition analyses were carried out at the National Food Safety Laboratory, Institute of Public Health, Dhaka, Bangladesh, accredited under ISO/IEC 17025. The mineral composition, including iron, zinc, iodine, calcium, selenium, potassium, sodium, magnesium, phosphorus, and copper, was analyzed using inductively coupled plasma mass spectrometry (ICP-MS) with collision cell technology (Thermo X, Series 2; Thermo Scientific, Hemel Hempstead, UK) following digestion with 69% nitric acid (Aristar® analytical grade; VWR Chemicals, Poole, UK) in a microwave digester (MARS Xpress; CEM Microwave Technology Ltd., Buckingham, UK). To account for interference, the ICP-MS used 100% helium as the collision gas while operating in kinetic energy discrimination (KED) mode. As internal standards, 10 ppm of gallium and scandium were utilized (BDH Chemicals Ltd., Poole, UK). Recovery testing and calibration with approved reference standards were part of the method validation process, according to Sprague et al. (2020) (30).

Fat composition was analyzed using Gas-liquid chromatography (GLC) (30) at the Institute of Aquaculture, University of Stirling, United Kingdom. Using an ultra-turraxing technique in chloroform-methanol (2:1, v/v) with 0.01% (w/v) butylated hydroxytoluene (BHT) as an antioxidant, total lipids were extracted from homogenized samples using the Folch et al. (1957) method (31). Gravimetric weight determination was used for the lipids. Fatty Acid Methyl Esters (FAME) were produced by acid-catalyzed transesterification with a 1% (v/v) sulfuric acid solution in methanol and 1 mL of toluene at 50°C for 16 hours. Using a Fisons GC-8160 (Thermo Scientific, Milan, Italy) fitted with a ZB-wax column (Phenomenex, Cheshire, UK) and flame ionization detection, FAMES were separated by gas-liquid chromatography (GLC), with heptadecanoic acid (17:0) serving as the internal standard. To ensure the precision of fatty acid quantification, calibration was carried out using the Restek 20-FAME Marine Oil Standard (Thames Restek Ltd., Buckinghamshire, UK).

Vitamin E, A1, B12 (cobalamin), and D3 were among the vitamins analyzed. Under ISO17025 accreditation, they were measured using high-performance liquid chromatography (HPLC) (32).

Based on the nutritional composition of the RUFP as presented in **Table S1** and the reference value for each nutrient for children aged 6 to 24 months, the potential contribution of the produced powder to the daily-recommended allowance (RDA) was estimated. A 5 g portion of RUFP provides 25% of the WHO-recommended protein intake for children (6 to 23 months) (30). Additionally, 5 g of RUFP may provide 2.1% of daily fat requirements (25 g per day) (33). For 7–23-month-olds, the RUFPs provide 39–195% of vitamin B₁₂ (34) and 4.8% of vitamin D₃ (34). However, the RUFPs barely meet 1.2 to 1.4% and 0.1 to 0.2% of children's daily vitamin E (35) and A₁ (34) requirements, respectively.

For micronutrients and trace elements, 5 g of the RUFP may provide 42 and 62% RDAs of calcium (36) for 6–11 and 12–23-month-olds, respectively. In addition, it supplies 23 to 40% and 6.6 to 6.8% of the RDA for iron and zinc (36), respectively. It also contains iodine and selenium (34). Children may acquire 0.3, 12.7–17.8, and 8.9–14.3% of their omega-6 (37), omega-3 (35), and EPA plus DHA's RDAs (34) with 5 g of the anchovy (olua), sardine (chapila), and thryssa (faissa) fish powder mixture, respectively (**Supplementary Table S1**).

Intervention

The intervention group was given 5 g of RUFPP per day with their complementary food, while the control group received no intervention. The RUFPP was supplied to the respective households at a 7-day interval, and both study groups were observed for sixteen weeks. Assigned VHVs were selected from Cox's Bazar district to comprehend and communicate in the local dialect effectively. Leaflets, the local language, containing information on the use of RUFPPs in recipe preparation, were delivered to caregivers in the intervention group. Before the trial, a pilot study was conducted at a similar site (Pechardwip, Ramu, Cox's Bazar) to determine children's food preferences and intake frequency.

Socio-demographic assessment

Basic socio-demographic information about the study participants (age and gender) and their caregivers (age, education, family income, access to latrine, and sources of drinking water) included in the study has been classified based on the Bangladesh Bureau of Statistics (BBS) 2020 classification (36).

Dietary assessment

A set of validated food frequency questionnaire (FFQ) was administered based on prior studies (38, 39) To assess the subjects' daily approximate nutrient intake, habitual dietary habits, specifically regarding the consumption of various food items (i.e., fruit and vegetables, carbohydrates, protein, dairy, fats, and oils), and the extent to which food items included the RUFPP as intervention, daily approximate nutrient intake was collected by a single 24-hour dietary recall method for both pre- and post-intervention. The frequency of consuming 10 dietary groups over every 14 days (39) and the Individual Dietary Diversity Score (IDDS) (40) have been calculated from the FFQ section for caregivers at a three-day interval.

Intervention monitoring and adherence assessment

Information was gathered during the intervention period. Two skillful VHVs collected data from every household at 3-day intervals, for a total of 39 visits during the 116-day study period. Each VHV was provided with an electronic tablet used to collect real-time information, images of prepared food, and household consumption patterns from households in the intervention group throughout the experimental period. The responses were duly recorded in the monitoring sheets. At the final stage of the study, all caregivers in the intervention group were assessed regarding their overall sensory and marketing preferences.

Outcome measures

Anthropometric outcomes

Measurements of children's Mid-Upper Arm Circumference (MUAC) using the Ibis Medical MUAC for the children and head circumference using the Seca head circumference measurement instrument (Model 2121817009, USA) were carried out at the pre- and post-intervention. The Camry Electronic Personal Scale (Model EB9300, China) was used to weigh infants wearing few or no outfits. Lengths were measured using a portable UNICEF wooden child measuring length board (precision 0.2 cm). All anthropometric information for infants was entered into the WHO Anthro Version 3.2.2 software. Weight and length were used to compute z-scores for weight-for-age (WAZ), length-for-age (LAZ), and weight-for-length (WLZ) using WHO thresholds (24). For WAZ, LAZ, and WLZ, z-scores of -2 to -3 standard deviations

(SD) below the study participants' means have been considered underweight, stunted, and wasted, respectively. In contrast, the values less than - 3 SD are considered severely underweight, severely stunted, and severely wasted (41). The length (69–86.8 cm for males and 67.3–85.1 cm for females) and weights (8.3–11.9 kg for males and 7.9 – 11.8 kg for females) (42), MUAC (< 11.5 → Severe malnutrition, 11.5–12.5 → Moderate malnutrition, > 12.5 → Normal) (43), and head circumference (43.3–48.3 cm for males and 42.8–47.2 cm for females) (44) were also compared with normal international standards.

Biochemical outcomes

To assess changes in various biomarkers following the intervention with RUFPs for the given period, 3 mL venous blood samples from children in the control and intervention groups were collected using BD Vacutainer SST II Advance tubes (without anticoagulant) at pre- and post-intervention time points. After collecting blood, random blood sugar (RBS) and hemoglobin (Hb) were measured immediately in uncoagulated full blood by a portable Glucometer (Accu-Chek, Roche, Germany) and a Hemoglobin meter (CERA-CHEK HB Plus). The blood sample was set to coagulate for 30 min, then centrifuged (10 min, 1000g, 20°C). Serum was collected and temporarily stored in an icebox (2–5 minutes) before freezing at -70°C pending analysis. The samples were analyzed at the biochemical laboratory of the International Centre for Diarrheal Disease Research, Bangladesh (ICDDR, B). The results obtained, retinol (µg/dl), 20 to 50 (45), vitamin B12 (pg/mL) 190 to 950 (46), ferritin (ng/mL) 7 to 150 (47), calcium (mg/dl) 8.8 to 10.8 (48), iron (µg/dl) 22 to 184 (49), Hb (g/dL) 11 to 13.5 (49), and RBS (mmol/L) 3.9 to 5.6 (50) were compared with standard reference values.

Caregivers' acceptability and marketing preferences

Caregivers' preferences regarding sensory and marketing indicators were assessed using a semi-structured questionnaire that included the hedonic scale. This scale, a symmetrical bipolar model with a neutral center, uses categories expressing varying levels of preference (51, 52). The food industry has made substantial use of penalty assessment (PA) using this hedonic scale (53). The following scores were assigned: 1 for 'very bad' or 'strongly disagree'; 2 for 'bad' or 'disagree'; 3 for 'neutral'; 4 for 'good' or 'agree'; and 5 for 'very good' or 'strongly agree' (54).

Statistical analyses

All data were entered into Microsoft Excel (version 2019) for coding and to ensure that the entered data were normal. The data were manually rechecked for missing values and converted to suitable file formats before being exported to SPSS (version 26) and RStudio (version 4.4.1). Descriptive data is given as mean (SD) or frequency (n) with percentages (%). To compare continuous variables (age, family size, monthly household income, hemoglobin, calcium, and RBS) between the intervention and control groups at both pre- and post-intervention, a one-way ANOVA was used. A chi-square test was employed to analyze categorical variables. A paired-samples t-test was employed to determine within-group differences in anthropometric and biomarker indicators at pre- and post-intervention. An independent-samples t-test was employed to determine the between-group difference in anthropometric and biomarker indicators at pre- and post-intervention. Lucid charts were used for the SWOT analysis. RStudio was used to represent consumption preferences with doughnut charts and biomarker changes with boxplots.

Results

Socio-demographic characteristics of the study population

The socioeconomic and demographic details of the children and their caregivers are shown in Table 1. Participant demographics were generally comparable between the control and intervention groups. Most caregivers were housewives with primary education, and over 70% of households relied on their own tube wells for water. Access to

improved latrines was around 40% across both groups. Family size was slightly smaller in the intervention group, while monthly household income tended to be higher. No significant differences were observed in children's or caregivers' age and caregiver education ($p < 0.05$).

Table 1
Socio-demographic characteristics of the study population by the group

Characteristic	No., N	Control	No., N	Intervention	p
Male children, n (%)	27	14 (52)	30	12 (40)	.370
Female children, n (%)		13 (48)		18 (60)	
Children's age (m), mean (SD)	27	13.8 (4.4)	30	12.1 (3.7)	.112
Caregiver age (y), mean (SD)	27	24 (4.2)	30	25.8 (6.2)	.225
Education of caregivers	Illiterate, n (%)	9 (33.3)	30	6 (20.0)	.196
	Primary education, n (%)	16 (59.3)		17 (56.7)	
	Secondary education, n (%)	2 (7.4)		7 (23.3)	
Sources of drinking water	Own tube-well, n (%)	21 (77.8)	30	21 (70.0)	.506
	Other tube-well, n (%)	6 (22.2)		9 (30)	
Access to a latrine	Improved, n (%)	11 (41)	30	12 (40)	.955
	Unimproved, n (%)	16 (59)		18 (60)	
Family size (n), mean (SD)	27	5.9 (2)	30	4.7 (2)	.038
Monthly household income (USD*), mean (SD)	27	109.0 $\pm$ 46.3	30	123.7 $\pm$ 62.1	.344

Here, values indicate mean (SD) or n (%). One-way ANOVA was conducted for continuous variables, and the Chi-square test was conducted for categorical variables.

* 1 USD = 106 BDT (Source: Bangladesh Bank, November 24, 2022)

** Indicates significant differences observed between the control and intervention groups ($P < 0.05$).

Biomarkers outcome

Table 2 illustrates the changes in biomarker levels from pre- to post-intervention among children in the control and intervention groups. Retinol concentrations increased in both groups, with a statistically significant increase observed only in the intervention group ($p < 0.001$). Ferritin levels showed greater improvement in the intervention group than in the control group, with a significant difference in the change between groups ($p = 0.030$), indicating improved iron storage status following the intervention.

Changes in vitamin B12 and serum iron levels were not statistically significant within or between groups ($p > 0.05$). Hemoglobin levels decreased significantly in the control group ($p < 0.001$) but increased significantly in the intervention group ($p = 0.002$); however, the difference in hemoglobin changes between groups was not statistically significant ($p = 0.320$).

Serum calcium increased significantly only in the control group ($p = 0.008$), whereas random blood sugar (RBS) levels decreased significantly only in the intervention group ($p = 0.005$). However, neither calcium nor RBS showed significant

differences in the magnitude of change between the control and intervention groups ($p > 0.05$) (see also Supplementary **Table S3**).

[For biomarker's percent increase or decrease, see supplementary **Table S4**]

Table 2
Change in biomarker measures from pre- and post-intervention for children

Biomarkers	Control Group					Intervention Group					p^b
	n	Pre-Mean (SD)	Post Mean (SD)	Change Mean (SD)	p^a	n	Pre-Mean (SD)	Post Mean (SD)	Change Mean (SD)	p^a	
Retinol (µg/dl)	9	24.9 (4.0)	29.8 (6.3)	4.9 (8.3)	.121	7	25.9 (3.5)	35.2 (2.2)	9.2 (2.2)	.000*	.218
Vitamin B12 (pg/ml)	8	497.8 (186.0)	489.9 (115.5)	- 7.8 (121.9)	.860	7	450.0 (248.1)	483.5 (226.1)	33.5 (65.1)	.223	.438
Ferritin (ng/ml)	8	23.4 (13.1)	22.1 (12.1)	- 1.3 (6.3)	.566	7	34.7 (26.4)	50.2 (42.8)	15.5 (18.4)	.068	.030*
Iron (µg/dl)	8	27.5 (7.8)	25.3 (10.2)	- 2.2 (9.8)	.541	7	33.9 (17.7)	37.5 (28.9)	3.6 (12)	.458	.320
Hb Level (g/dl)	17	12.2 (1.1)	11.2 (1.5)	- 1 (.8)	.000*	19	11.9 (1.4)	12.9 (.8)	1 (1.3)	.002*	.320
Calcium (mg/dl)	8	9.9 (0.4)	10.5 (0.6)	.6 (.5)	.008*	7	10.2 (0.4)	10.5 (0.5)	.3 (.7)	.346	.741
RBS Level (mmol/L)	15	6 (0.5)	5.7 (0.5)	- .3 (.6)	.097	19	5.4 (0.4)	5 (0.5)	- .4 (.5)	.005*	.641

p^a for comparison within the groups (pre vs post), paired samples t-test.

p^b for comparing the groups (control vs intervention): an independent-samples t-test.

*Indicates significant differences observed at a p-value of < 0.05 .

Anthropometric outcome

Table 3 baseline anthropometric indicators were comparable across groups. Both the control and intervention groups demonstrated significant increases in body length ($p < 0.05$), reflecting normal growth with age. However, length-for-age z-scores (LAZ) decreased significantly in both groups (control: $p = 0.013$; intervention: $p = 0.003$), indicating that although children grew in absolute length, their linear growth velocity was lower than expected for age- and sex-specific growth standards. However, between-group differences were not significant for any indicators ($p > 0.05$). Mid-upper arm circumference (MUAC) displayed a positive but non-significant ($p > 0.05$) rise in both groups (see also Supplementary **Table S4**).

Table 3
Anthropometric outcomes from the beginning to the end of the study for children aged 6 to 23 months

Anthropometric Indicators	Control Group (N = 19)					Intervention Group (N = 21)					p ^b
	No., n	Pre-Mean (SD)	Post Mean (SD)	Change Mean (SD)	p ^a	No., n	Pre-Mean (SD)	Post Mean (SD)	Change Mean (SD)	p ^a	
Body weight (kg)	19	9.1 (1.6)	10.3 (1.5)	1.2 (.8)	.000*	21	8.2 (1.3)	9.4 (1.1)	1.2 (.7)	.000*	.908
Body length (cm)	19	74.4 (6.9)	76.7 (7.5)	2.4 (2.8)	.002*	21	71.4 (5.5)	73.5 (5.4)	2.1 (2.5)	.001*	.768
MUAC (cm)	19	143.3 (12)	148.7 (16.6)	5.4 (13.5)	.099	21	141.9 (10.7)	144.7 (7.9)	2.8 (11.5)	.275	.520
Head circumference (cm)	19	44.7 (2.5)	45.7 (2.2)	1.0 (1.4)	.006*	21	43.6 (1.7)	44.6 (1.4)	.9 (1.1)	.001*	.807
Weight-for-age z-score (WAZ)	19	-0.6 (1.2)	-0.3 (0.9)	0.3 (0.8)	.098	21	-1.0 (1.1)	-0.6 (0.8)	0.5 (.7)	.006*	.521
Length-for-age z-score (LAZ)	19	-0.9 (1.4)	-1.6 (1.7)	-.7 (1.1)	.013*	21	-1.2 (.9)	-1.9 (1.1)	-0.7 (1)	.003*	.943
Weight-for-length z-score (WLZ)	19	-0.1 (1.3)	0.8 (1.2)	.9 (1.3)	.012*	21	-0.5 (1.2)	0.5 (1)	1 (.7)	.000*	.617

p^a for comparison within the groups (pre vs post), paired samples t-test.

p^b for comparison between the groups (control vs intervention), an independent-samples t-test.

*Indicates significant differences observed at a p-value of < 0.05.

Consumption scenario - dietary intake

When comparing the IDDS values for the two groups to the maximum IDDS value of eight, the magnitudes are noticeably lower for both groups. The IDDS score indicates a dietary pattern with lower diversity and lower nutritional richness among participants in both the control and intervention groups, as their scores were almost identical. It is evident from the consumption patterns and IDDS score that the members of this particular group had almost identical scores, indicating a dietary pattern with less variation and lower nutritional richness (See supplementary **Figures S1** and **S2**). Usual macro- and micronutrient intake, assessed by 24-hour dietary recall, is summarized in Table S2. Mean nutrient intakes and their corresponding percentage contributions to the daily RDA are presented separately for the control and intervention groups across macronutrients, vitamins, and minerals.

Compliance with the intervention

Compliance with the intervention protocol was generally high throughout the four-month (116-day) study period. Among intervention households, 97% supplemented their children's meals with fish powder for more than 80%. Moreover, 65% of the target consumers consumed fish-powder supplements on more than 80% of the monitored days, whereas 32% consumed them on 60–80% of the monitored days. Caregiver-reported feeding behavior showed good acceptance of the intervention foods. Approximately 37% of the enrolled children consumed intervention foods at a greater rate than other foods, and 36% of the respondents claimed that children consumed approximately the same

amount of food as the rest of the family. Additionally, just 27% of the intervention group consumed a small amount of food (**Figure S3**).

Whereas **Figure S4** depicts the types of foods and food preferences for consumption of the combined powder of pelagic small fish. According to the caregiver's responses, the participants preferred khichuri, noodles, vegetables, and legumes to other foods. These foods were also the most well-liked by children and the adults who looked after them.

Choice of caregivers'

The organoleptic assessment was done by giving each of the three attributes—color, smell, and taste—a 5-point rating (**Figure S5**). The scenario was almost identical, except for color. 44% of respondents gave the smell a favorable rating (score 4), and 40% rated it excellent (score 5). Similarly, 40% of respondents said the taste of products made with fish powder was good or very good; only 20% were neutral. However, 44% of study respondents rated the fish powder "not very good" (score 1), while 40% rated it "not good" (score 2). Furthermore, 16% of respondents gave neutral feedback on the color of the RUFs.

84% of survey participants strongly agreed that fish powder-based food products are safer than dried and raw fish. it. After eating, very few respondents (12%) reported stomach discomfort. A sizable portion (92%) of respondents believe that fish powder is less likely to be tampered with.

According to 64% of respondents, the taste of fish powder cannot be altered once it has been preserved. 96% of respondents believe it may improve children's health, and 76% of respondents believe that fish powder-based food items have a higher nutritional value than other fish products. 40% of respondents strongly agreed that meal items made with fish powder are more nutritious than cooked foods. 96% of respondents believe that fish powder-based food items can provide significant nutrition to the impoverished community.

Market acceptability and perceptions

Caregivers' preferences regarding fish powder-based products are brief in **Table S5**. Overall, 84% of respondents stated a strong interest in buying fish powder products and recommending them to others. Most favored buying such products from local markets (40%) and from reputable companies or providers (84%), with 80% demonstrating willingness to purchase year-round. The favored package size ranged from 50 to 150 grams, priced between USD 0.5 and 1.75.

A summary of the SWOT (Strengths, Weaknesses, Opportunities, and Threats) analysis highlights that caregivers perceived the product's main strengths to be its ease of use, potential contribution to children's health, reduced cooking time, and high protein content. Besides, no major intimidations were identified with the products (**Figure S6**).

Discussion

This study investigated the effect of a ready-to-use fish-based complementary food product (RFUP) on biochemical and anthropometric indicators of nutritional status among children aged 6–23 months. The findings demonstrated that RFUP consumption was associated with favorable changes in selected micronutrient biomarkers, particularly a significant improvement in vitamin A status, as reflected by increased retinol concentrations, and improved iron stores, as indicated by the significant between-group increase in ferritin levels. Hemoglobin concentrations also increased significantly within the intervention group, although the difference in change between groups was not statistically significant. No significant intervention effects were observed for vitamin B12, serum iron, or anthropometric indicators.

The experimental group exhibited a significant increase in retinol concentrations throughout this study. A similar observation has also been reported for vitamin A levels in two distinct randomized controlled trials (RCTs) after the implementation of interventions, including fortified wheat flour chapatti (55) and fortified *Moringa oleifera* porridge (56), among children in the experimental group. However, additional randomized research on moringa leaf powder (57) also observed a rise in retinol levels, but the increase did not reach statistical significance. It is conceivable that the levels of retinol, a constituent of the vitamin A group, may undergo alterations as a result of a modest increase (19.9% vs. 35.6%) in the contribution to the recommended daily allowance (RDA) of retinol from both normal dietary intake and the incorporation of composite fish powder (**Supplementary Tables S1 and S2**).

The increase in vitamin B12 levels across the groups was slight (-7.8 vs 33.5), although the changes were not statistically significant. A randomized controlled trial conducted in Burkina Faso found that the use of Ready-to-Use Therapeutic Food (RUTF) for severe acute malnutrition (SAM) improved children's B12 status. However, it is important to note that the intervention did not fully correct the B12 deficiency in these children (58).

The intervention led to a favorable effect on iron status. A significantly greater change in ferritin shows this compared to the control group. However, the increase in ferritin in the intervention group was not statistically significant. Serum iron concentrations remained unchanged across the groups. Hemoglobin improved significantly in the intervention group, but the difference between groups was not statistically significant. A fish sauce intervention study among school children in Cambodia also showed a significant increase in serum ferritin and hemoglobin levels (59). Two other trials among children have similarly shown statistically significant improvements in hemoglobin levels within the intervention group (60, 61). Research examining a ready-to-use diet made from soya, maize, and sorghum (61), as well as another RCT involving children aged 6–59 months with severe acute malnutrition (SAM) (63), both showed a noteworthy increase in iron levels among the children. Notably, a study conducted in Kenya utilized a sodium iron edetic acid (NaFeEDTA) approach, which resulted in enhancements across all iron-related biomarkers in children (62). Conversely, a study conducted in Cambodia among children aged 6–59 months found no significant difference in hemoglobin or ferritin levels between admission and discharge (63). While there was no substantial rise in iron levels in the intervention group, it is worth noting that an increase in ferritin levels is crucial for maintaining intracellular iron balance (64). The proximate iron consumption from the regular diet of both study groups was generally similar (6.5–11% vs. 8.5–14.4% of daily RDA); in this case, RUFPS iron (23–40% of daily RDA) potentially exerted the most impact on serum iron-related markers (**Supplementary Tables S1 and S2**). Based on existing data, a positive association between dietary iron intake and ferritin and hemoglobin levels has been shown (65, 66).

The study population consisted of individuals with normal blood calcium levels, specifically 8.8–10.8 mg/dl. Both groups showed an increase in these levels, with a slightly greater increase in the control group (.6 vs. .3 mg/dl). However, statistical analysis revealed that these differences were not statistically significant. This may be interpreted as showing that the serum calcium was not positively affected by the study product. Many dietary components, including oxalate, phytate, and tannin, may inhibit absorption and may be responsible for the lack of a significant impact of the study product on serum calcium (67, 68). Additionally, dietary protein and salt (69) and vitamin D (57) may influence the absorption of dietary calcium.

Although RBS levels decreased in both groups, the reduction was significant only in the intervention group. The magnitude of the reduction was slightly greater in the intervention group (-.3 mmol/L) than in the control group (-.4 mmol/L); however, the difference in change between groups was not statistically significant. Therefore, the observed reduction cannot be directly attributed to RFUP consumption. Nevertheless, the reduction in RBS in the intervention group may warrant further investigation, as dietary components such as protein and vitamin D have been implicated in glucose metabolism and insulin regulation in previous studies (70–72).

According to the above parameters, fish powder-based RUFs may enhance nutritional biomarkers in children under 2 years of age, but with mixed effects on anthropometric outcomes. Although absolute body length increased during the intervention, length-for-age z-scores (LAZ) decreased significantly in both groups with the same magnitude, indicating that children's linear growth did not keep pace with age-related growth expectations. In contrast, a 10-month Zambian linear mixed-effects model (21) The study revealed that fish powder enhanced linear growth in children aged 6 months and older throughout the intervention period. In this research, the fish powder was observed to increase length-for-age z scores by 1.3 (95% confidence interval [CI]: 0.94–1.57) and weight-for-age z scores by 1.0 (95% confidence interval [CI]: 0.6–1.25). The positive benefits of dietary intervention or supplementation on children's growth and development have also been demonstrated in a 10-month randomized controlled trial in Madagascar (73) and a meta-analysis of 79 RCTs (74). However, comparable research conducted in Cambodia did not discover any correlation between the endpoint and the baseline following the intervention using locally made ready-to-use supplemental foods on anthropometric indicators (24).

Policy implications and conclusion

The findings of this study highlight the potential of fish-based complementary foods as a targeted nutritional strategy to support young children in vulnerable fishing communities during periods of seasonal food insecurity, such as fishing bans. Although current government support programs, including rice distribution, may improve household food availability, they may not fully meet young children's micronutrient requirements. The observed improvements in selected micronutrient biomarkers, particularly retinol and ferritin status, suggest that nutrient-dense fish-based products may complement existing food assistance programs. However, further studies are needed to evaluate the feasibility, acceptability, cost-effectiveness, and scalability of integrating such products into broader social protection and nutrition programs.

Similar challenges may be experienced by other coastal communities facing seasonal livelihood restrictions. Therefore, the findings provide preliminary evidence that may inform future exploration of locally produced, nutrient-rich food solutions in comparable settings.

Strength and limitations

A key strength of this study is the evaluation of a locally developed fish-based complementary food intervention in a real-world community setting among vulnerable coastal fishing households in Bangladesh, enhancing the practical relevance and potential applicability of the findings. Furthermore, the inclusion of biochemical biomarkers alongside anthropometric measurements provided a more comprehensive and objective assessment of the intervention's effects on child nutritional status.

Nevertheless, several limitations should be considered. The study was conducted in a community setting, where practical constraints limited the ability to implement a fully rigorous randomized controlled trial design, potentially introducing sources of bias. The relatively small sample size, largely due to funding constraints, may have limited the statistical power to detect modest intervention effects, particularly regarding growth outcomes. Finally, the short duration of the intervention and follow-up restricted the assessment of long-term effects on child growth and nutritional status. Finally, although the RFUP provided multiple nutrients, including zinc, iodine, selenium, and n-3 polyunsaturated fatty acids, the biochemical assessment was limited to selected markers, preventing a full evaluation of the intervention's effects on overall micronutrient and fatty acid status.

Conclusion

This study demonstrates that supplementation with a locally developed fish-based complementary food product may improve selected micronutrient indicators among children aged 6–23 months living in vulnerable coastal fishing communities in Bangladesh. Specifically, the intervention was associated with improved vitamin A status, reflected by increased retinol concentrations, and favorable changes in iron status, as indicated by a significantly greater increase in serum ferritin than in the control group. Although hemoglobin concentrations increased significantly within the intervention group, no significant between-group differences were observed for hemoglobin, vitamin B12, serum iron, or anthropometric indicators.

These findings suggest that nutrient-dense fish-based complementary foods may be a promising, locally relevant strategy to support the nutritional needs of young children during periods of seasonal food insecurity, such as during fishing bans. However, given the small sample size, short intervention period, and community-based study design, larger, longer-term studies are needed to confirm these findings and assess the feasibility, effectiveness, and scalability of integrating these interventions into broader nutrition and social protection programs.

Abbreviations

AOAC	Association of Official Analytical Chemists
BDHS	Bangladesh Demographic and Health Survey
DHA	docosahexaenoic acid
EPA	eicosapentaenoic acid
FFQ	food frequency questionnaire
Hb	hemoglobin
IDDS	Individual Dietary Diversity Score
IYCF	Infant and Young Child Feeding
LAZ	length-for-age z-score
MUAC	Mid-Upper Arm Circumference
n	3 PUFA-long-chain n-3 polyunsaturated fatty acids
PA	penalty assessment
PLW	pregnant and lactating women
RBS	random blood sugar
RDA	recommended daily allowance

RUFP
ready-to-use fish product
VHV
village health volunteers
WAZ
weight-for-age z-score
WLZ
weight-for-length z-score

Declarations

Ethics approval and consent to participate:

The Noakhali Science and Technology University Ethical Committee (NSTUEC) approved the research project after receiving written informed consent from each participant (Reg: NSTU/SCJ/EC/2021/73). Additionally, written content was obtained from each child's caregiver. All procedures involving human participants were performed in accordance with the ethical standards of the institutional research committee and with the 1964 Declaration of Helsinki and its later amendments.

Consent for publication:

Not applicable.

Competing interests:

There was no competing interest in conducting this research.

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Author Contribution

Abdullah-Al Mamuna: Conceptualization, Methodology, Supervision, Validation, Writing- Reviewing and Editing, Project administration, Funding acquisition, Resources. Shahedul Islam: Software, Data curation, Formal analysis, Visualization, Software, Writing- Original draft preparation, Writing- Reviewing and Editing. MD Nahiduzzaman: Writing- Reviewing and Editing. Samiul Islam: Validation, Writing- Reviewing and Editing. MA Rifat: Validation, Writing- Reviewing and Editing. Oumma Halima: Formal analysis, Software, Resources. A.B.M. Mahfuzul Haque: Validation, Writing- Reviewing and Editing. Muhammad Arifur Rahman: Data curation, Investigation, Resources. Abdul Wahab: Conceptualization, Validation, Writing- Reviewing and Editing. Shakuntala Haraksingh Thilsted: Conceptualization, Methodology, Validation, Writing- Reviewing and Editing. Marian Kjellefold: Conceptualization, Methodology, Validation, Writing- Reviewing and Editing, Resources. Maria Wik Markhus: Methodology, Validation, Writing- Reviewing and Editing, Resources.

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Data Availability

Availability of data and materials: The datasets analysed during the current study are available from the corresponding author on reasonable request.

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Figures

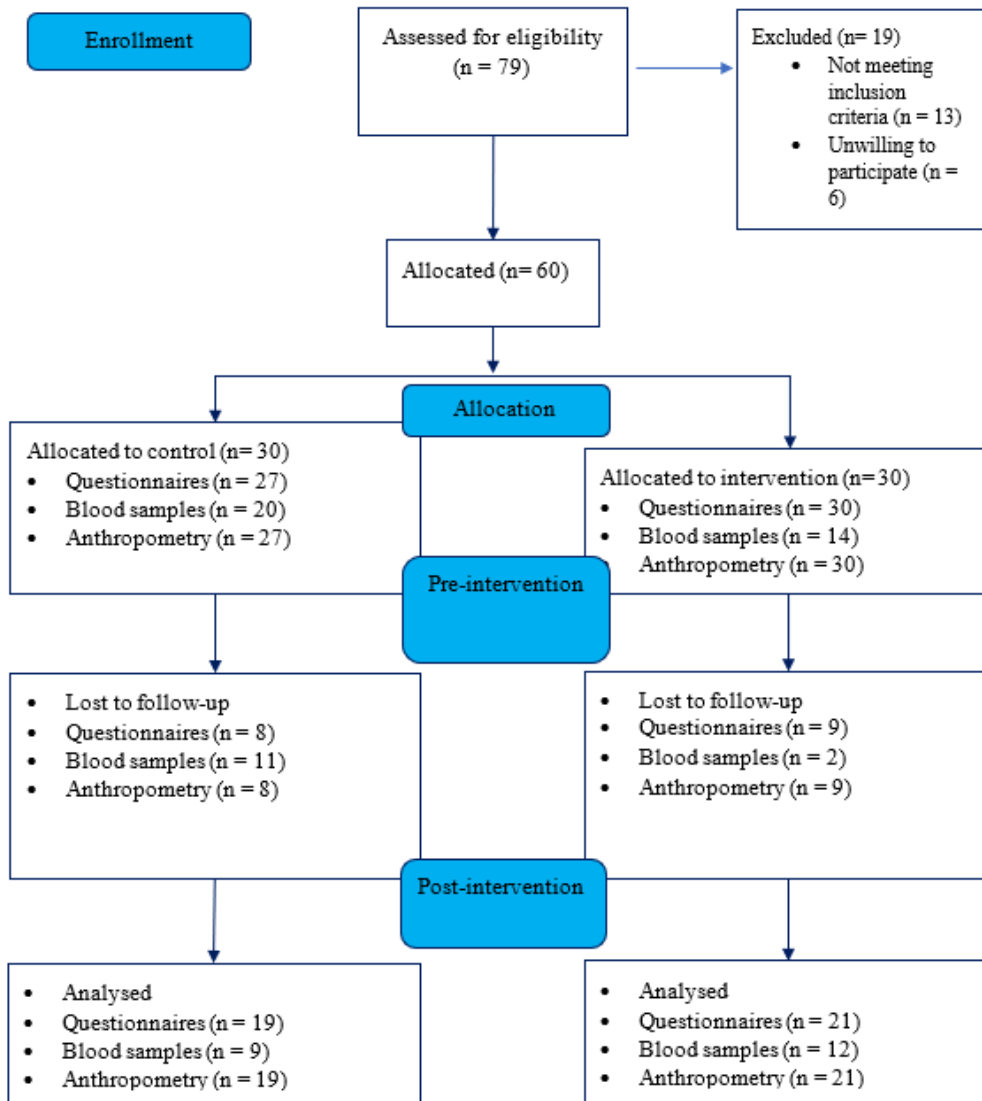


Figure 1

Flow diagram of the study participants.

Supplementary Files

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- [SupplementaryFile1.docx](#)
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