Original article

Impact of iron and zinc supplementation on child disability screening using the Ten Questions Plus tool in rural Sarlahi District, southern Nepal

Wu LA[1] , Katz J[2] , Mullany LC[2], Khatry SK[3], Darmstadt GL[2,4], LeClerq SC[2,3], Tielsch JM[5]

[1] Policy Advisor, National Vaccine Program Office, U.S. Department of Health and Human Services, 200 Independence Ave. SW, Washington, DC, USA; [2] Department of International Health, Bloomberg School of Public Health, Johns Hopkins University, 615 N. Wolfe Street, E5527, Baltimore, Maryland, [3] Nepal Nutrition Intervention Project, Sarlahi (NNIPS), Kathmandu, Nepal; [4] Director, Family Health Division, Global Health Program, Bill and Melinda Gates Foundation, P.O. Box 23350, Seattle, WA, USA. [5] Department of Global Health, George Washington University School of Public Health and Health Servicesl. 2175 K. Street NW Suite 200. Washington DC, USA.

Correspondence: Prof. Joanne Katz,

Email: jkatz1@jhu.edu

Abstract

Introduction: The Ten Questions tool was developed in 1984 as a low-cost tool to screen for child disability prior to referral for definitive diagnosis. Iron and zinc contribute to child growth and development, but few studies examine the relationship between supplementation and longer-term neuro-developmental outcomes.

Methods: The Ten Questions Plus tool was administered to caregivers of 1410 children between 1-9 years of age from August 2007 to March 2008 in rural Sarlahi, southern Nepal. Participants had been previously enrolled in a randomized 2x2 factorial trial of daily iron and zinc supplementation between October 2001 and January 2006.

Results: Nearly 30% of the sample screened positive for disability. The most common problems were learning disability (12.3%) and motor milestone delay (11.3%). Children supplemented with zinc were less likely to screen positive overall, and for motor and learning problems. Children supplemented with smaller doses of iron were more likely to screen positive overall, and for physical, and cognitive problems, but these differences were not statistically significant. There were no differences in children dually supplemented with iron and zinc compared with placebo.

Conclusions: The Ten Questions Plus tool can be used in this rural Nepali setting to identify children at increased risk for physical and cognitive disability who should be referred for clinical examination, diagnosis and enrollment in intervention services. We found evidence for a beneficial effect of zinc supplementation in reducing the likelihood of screening positive for disability.

Key Words: micronutrient supplementation, zinc, iron, disability, children, Nepal, screening

Introduction

The United Nations Children's Fund (UNICEF) estimates that 200 million children under five years of age in developing countries are not reaching their developmental potential[1]. These children are likely to suffer increased incidence of infections such as pneumonia and diarrhea, and do poorly in school, further putting them at risk for future lower income and continuing the cycle of poverty in later generations[2]. The greatest number of under-fives not meeting their developmental potential are in South Asia (an estimated 88.8 million, 52% of all under-fives in the region)[3]. A complicated interplay of genetic, nutritional, environmental and social factors, including iron and other micronutrient status, may contribute to a child's development, and studies of the effects of these factors can help us better understand how to optimize neuro-developmental and physical growth processes[4].

Micronutrient supplementation with iron and zinc promotes growth of the brain, thus affecting sensory-motor, cognitive, and social-emotional development[1,4]. Iron supplementation trials have shown benefits for motor, social-emotional, and cognitive-language outcomes. Because these trials only followed children up to 12 months, little is known about the long-term effects of iron supplementation on physical and cognitive development[4-6]. Zinc supplementation trials in infants and young children in developing country settings have shown reduced mortality, diarrhea and pneumonia morbidity, and improved growth[7-9]. Although the effects of individual iron or zinc supplementation in children on mortality and specific morbidities in developing country settings have been extensively studied, neuro-developmental outcomes have been less frequently examined[8,10]. As efforts to address under-five mortality have led to global improvements in the delivery of existing effective interventions to mothers and children in low income countries, more children will survive their early years and enter school[11-13].

In developing countries, widespread diagnoses of disability are not typically available due to limited resources, poor health systems and other health priorities. What is generally needed is a process for screening for neuro-developmental disabilities at population or primary care level, followed by referral for definitive diagnosis and intervention services. Early diagnosis and intervention are important to target children who need further medical attention, and to optimize the chances to mitigate the degree of impairment[14].

The Ten Questions (TQ) questionnaire was developed in 1984 to screen children aged 2-9 years in resource-poor settings to identify disabilities and to refer children who screen positive for further clinical evaluation and intervention[15-16]. The tool focuses on questions about general functional abilities and milestones, rather than culture-specific skills[17]. Parents are asked about motor milestone delay, learning, vision, hearing, speech and physical problems in their child; a child is considered screen-positive if his or her parent responds 'yes' to one or more of the ten questions. The TQ has been globally validated in over 20 countries and is highly sensitive[18-20]. While the tool has a low positive predictive value for serious disability, these "false-positive" children are often found to have either mild disability or a health condition requiring treatment[19]. Thus, the TQ is best used to identify children with neuro-developmental delays who may be at increased risk of disability or impairment[1].

An additional question has been added to the TQ, assessing behavioral problems (e.g., frequent tantrums, aggressive behavior, and difficulty relating to people)[21]. The extended tool, referred to as the TQ Plus (TQP), was validated in Bangladesh. In another TQ study, the term "Ten Questions Plus" was used to refer to the TQ with the addition of direct testing of vision and hearing to supplement caregiver report[18].

Given the lack of data on the effects of iron and zinc supplementation on child disability in developing country settings, our objective was to examine the impact of iron and zinc supplementation on screen positivity for childhood disability using the Ten Questions Plus tool in rural southern Nepal. We analyzed both the intent-to-treat (randomized micronutrient supplementation group) and dose-response effects (number of iron and zinc tablets taken) in a subsample of children who had participated in a larger randomized trial of iron and zinc supplementation[22,23]. We also assessed the associations of micronutrient supplementation with positive responses to individual questions to examine particular types of reported disability (e.g., motor/physical, cognitive, vision, and hearing).

Methods

The TQP (including behavioral testing) was administered to a sample of children as part of a follow-up in the Nepal Nutrition Intervention Project, Sarlahi (NNIPS-4), a community-based, cluster-randomized 2x2 factorial trial of the effect of daily iron and/or zinc supplementation on mortality, morbidity, and physical and mental development in children 1-36 months old[22-23]. Children were randomized to receive one of four oral supplements daily: iron (12.5 mg) and folic acid (50 µg); zinc (10 mg); iron, folic acid, and zinc; or placebo. Children 1-11 months of age received half the dose. All children were visited twice every week; study staff directly observed the ingestion of the tablets on those days. Tablets for daily doses until the next visit were left with the child's caregiver. Older children took the tablets directly and caregivers were instructed to dissolve the tablets in clean water or breast milk for younger children. During the visits, compliance with supplementation was assessed, and the number of tablets consumed in the preceding week was recorded for each child. All investigators, study staff, and participants were blinded to the assigned treatment.

NNIPS-4 was conducted in Sarlahi District between October 2001 and January 2006 among 426 randomized clusters within 30 Village Development Committees (VDC), a government-defined administrative unit. Sarlahi District is in the low-lying plains of Nepal, bordering the Indian state of Bihar. The residents in this area are typically subsistence farmers or laborers, characterized by traditional rural Hindu culture, and have lower household wealth relative to other areas in Nepal[22,24]. Household characteristics and socioeconomic status such as caste, ethnicity, parental literacy, and ownership of various items indicating household wealth were recorded at enrolment into the trial.

The iron arms of NNIPS-4 were stopped in November 2003 upon the recommendation of the data safety and monitoring board (DSMB) due to lack of a mortality or morbidity effect of iron-folic acid[23]. Children who had been randomized to an iron-folic acid containing group and newborns were subsequently randomized to zinc only or placebo.

The sample for the current study included children residing in one of the 30 VDCs, Ishwarpur (comprising 22 clusters). Thus, while not a random sample of NNIPS-4 participants, the children selected for interview represent a geographically distributed population throughout Ishwarpur. The TQP instrument was translated from English to Nepali and then back translated by a different person to ensure the two versions were the same. TQP administration began in September 2007, and data were collected through March 2008. A summary indicator variable for positive disability screening was defined as a positive response to one or more of the eleven screening questions. We modeled the crude and adjusted odds of a positive screen, as well as a positive response on individual questions where there was a 2% or greater overall prevalence, as a function of treatment groups (intent-to-treat analysis with placebo being the reference). We also examined the associations as a function of the number of iron and zinc tablets taken (per-protocol analysis). Logistic regression models were adjusted for potential confounding variables found to be associated with the main covariate(s), the outcome, or both. Likelihood ratio tests to compare extended models to null models not adjusted for potential confounders were also analyzed. The number of unique covariate patterns in the data was examined, and we assessed goodness-of-fit of the model using Pearson's method because there were many non-unique covariate patterns. We also looked at model residuals, leverage, and influence to check model fit and assumptions. Data were analyzed using Stata 10 and SAS 9.2[25,26].

The study was reviewed and approved by the Nepal Health Research Council, Kathmandu, and the Institutional Review Board of the Johns Hopkins Bloomberg School of Public Health. The NNIPS-4 trial is registered at clinicaltrials.gov (NCT 00109551).

Results

There were 3194 children from Ishwarpur VDC in the NNIPS-4 micronutrient supplementation trial (Figure 1). Of these, 28 died, 59 moved, and 1 refused to participate in the TQP disability screening, leaving 3106 children eligible to receive TQP testing. Follow-up measurements (e.g., mortality and morbidities such as diarrheal disease and respiratory infections) on these children started in early 2007, andTQP administration began September 19, 2007, after interviewers had been appropriately trained. 1902 children were administered TQP testing. 492 of these 1902 children were originally randomized to an iron-folic acid containing supplementation group, and were re-randomized to either zinc or placebo supplements upon the recommendation of the DSMB[23]. These re-randomized children were not included in the current analysis, and the final sample includes 1410 children; the children in the iron-containing groups in our sample are those that aged out of the study before November 2003 and were not re-randomized, and were therefore not exposed to zinc supplements.

At the baseline of NNIPS-4, the age of study entry in the TQP subset ranged from 28 days to 2.8 years, and the age of study exit from NNIPS-4 ranged from 43 days to 3 years. The length of time in the study ranged from 13 days to 2.9 years with a mean of 1.5 years. 51.4% of the children were male. Age at the time of TQP administration ranged from 1.4 to 8.9 years. This population was malnourished with high proportions of wasting and stunting [39.1% weight-for-height Z score < -2 standard deviations from the mean (SD) (i.e., wasted) and 51.2% height-for-age Z score < -2 SD (i.e., stunted), respectively]. The majority of households were subsistence farmers with low parental literacy (roughly 52% for fathers and 20% for mothers). Baseline characteristics by supplementation group are shown in Table 1.

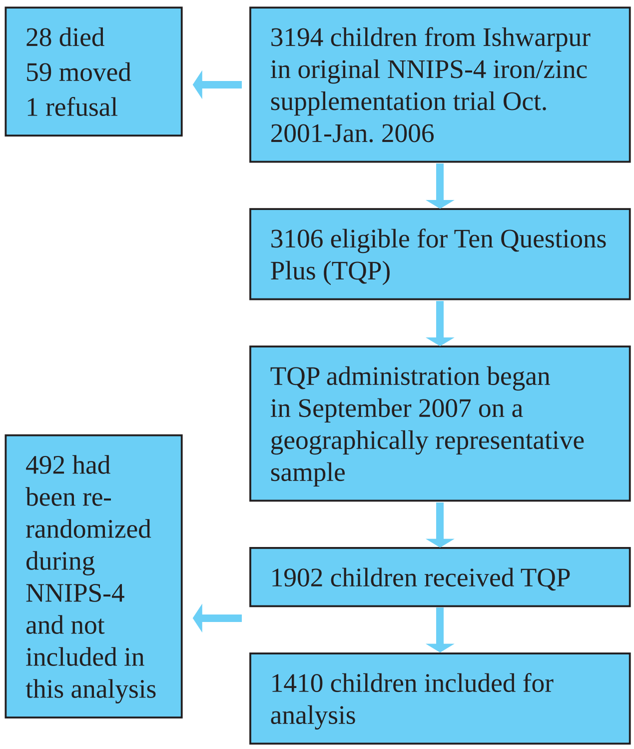

Figure 1 Study Profile on micronutrient supplementation trial in children

Table 1 Characteristics of Children Administered TQP from Ishwarpur VDC, N=1410

	Mean (SD) or n (%)			
	Placebo Group N=875	Zinc Only Group N=334	Iron Only Group N=77	Iron & Zinc Group N=124
Baseline child characteristics:				
Age of study entry (yrs)	0.4 (0.6)	0.4 (0.6)	1.8 (0.5)	1.8 (0.5)
Age of study exit (yrs)	2.4 (1.0)	1.9 (1.0)	2.9 (0.4)	2.9 (0.4)
Time in study (yrs)	1.6 (0.9)	1.5 (0.9)	1.1 (0.5)	1.04 (0.5)
Male	421 (48.1%)	162 (48.5%)	40 (52.0%)	62 (50.0%)
Child characteristics at TQP administration:				
Age (yrs)	4.5 (1.9)	4.2 (1.8)	7.6 (0.6)	7.7 (0.6)
WAZ score < -2 SD*	555 (73.6%)	173 (61.1%)	53 (74.7%)	72 (72.7%)
HAZ < -2 SD*	382 (54.7%)	107 (40.7%)	42 (59.2%)	49 (49.5%)
WHZ <- 2 SD*	263 (38.5%)	98 (38.9%)	22 (36.1%)	40 (46.5%)
BMIZ < -2 SD*	209 (29.9%)	90 (34.5%)	25 (35.2%)	44 (44.4%)
Baseline household characteristics:				
Caste				
Brahmin	41 (4.7%)	21 (6.3%)	3 (3.9%)	4 (3.2%)
Chhetri	13 (1.5%)	22 (6.7%)	0 (0.0%)	4 (3.2%)
Vaiysha	426 (48.7%)	238 (71.9%)	59 (76.6%)	73 (58.9%)
Shudra	124 (14.2%)	26 (7.9%)	7 (9.1%)	24 (19.4%)
Muslim	270 (39.0%)	24 (7.3%)	8 (10.4%)	19 (15.3%)
Ethnicity				
Pahadi	108 (12.4%)	87 (26.4%)	7 (9.1%)	16 (13.0%)
Madheshi	760 (87.6%)	242 (73.6%)	70 (90.9%)	107 (87.0%)
Mother literate	173 (20.4%)	83 (26.2%)	15 (19.5%)	20 (16.5%)
Father literate	438 (50.1%)	180 (53.9%)	45 (58.4%)	70 (56.5%)
Latrine in house	69 (7.9%)	21 (6.3%)	6 (7.8%)	20 (16.1%)
Servant(s) in house	37 (4.2%)	21 (6.3%)	7 (9.1%)	3 (2.4%)
Cattle	467 (53.4%)	201 (60.5%)	39 (50.7%)	53 (42.7%)
Goat(s)	356 (40.7%)	117 (35.2%)	32 (41.6%0	31 (25.0%)
Land ownership	372 (43.1%)	193 (59.9%)	38 (50.0%)	57 (46.0%)
Bullock cart(s)	95 (10.9%)	38 (11.5%)	9 (11.8%)	6 (4.8%)
Bicycle(s)	443 (50.7%)	185 (55.9%)	31 (40.8%)	57 (46.0%)
Radio(s)	185 (21.2%)	97 (29.2%)	16 (21.1%)	25 (20.2%)
Electricity in house	236 (27.0%)	95 (28.6%)	23 (29.9%)	37 (29.8%)
Television(s)	151 (17.4%)	85 (25.6%)	12 (15.8%)	33 (26.6%)

Abbreviations: SD = standard deviations from the mean, WAZ = weight for age Z score, HAZ = height for age Z score, WHZ = weight for height Z score, BMIZ = body mass index for age Z score

* Combination of WHO 2000 and 2005 reference populations, each of which covers different age range. 2000 reference population was used only if not in age range for 2005 reference population.

Missing values (of N=1410): WAZ: 203, HAZ: 278, WHZ: 327, BMIZ: 280, Caste: 4, Ethnicity: 13, Maternal literacy: 46, Latrine: 5, Servants: 3, Cattle: 3, Goats: 3, Land: 25, Carts: 4, Bicycles: 5, Radios: 4, Electricity: 3, Televisions: 8

TQP respondents were mothers (69.9%), fathers (5.1%), and other relatives (25.0%). Overall, 28.9% of children screened positive with a positive response to at least one question. The prevalence of positive responses on individual questions ranged from 0.9% for vision problems (Q2), to 12.3% for learning problems (Q7). There was a relatively high report of motor milestone delay (Q1) (11.3%). The prevalence of positive TQP responses by treatment group is shown in Table 2. 62.1% (N=875) of the sample were in the placebo supplementation group; 23.7% (N=334) were in the zinc only group; 5.5% (N=77) were in the iron only group; and 8.8% (N=124) were in the iron and zinc group. There were fewer children in the iron only and iron and zinc groups because the iron-containing arms were halted in November 2003 as described above. The number of iron tablets taken ranged from 0-594, with a mean of 30.0 [standard deviation (SD)=96.6]. The number of zinc tablets taken ranged from 0-1052, with a mean of 104.7 (SD=212.9).

Children in the zinc only group had reduced crude and adjusted prevalence of positive disability screening (adjusted (a) OR=0.70, 95% confidence interval (CI): 0.51-0.94) (Table 3). This effect was primarily driven by reduced relative odds of motor milestone delay (aOR=0.57, 95% CI: 0.32-0.81) and learning problems (aOR=0.56, 95% CI: 0.36-0.86). There was no statistical evidence of impact of iron and zinc combination treatment, or iron only treatment on screen positivity.

Table 2 Prevalence of Positive TQP Responses, N=1410

Outcome		**Placebo Group** N=875	**Zinc Only Group** N=334	**Iron Only Group** N=77	**Iron & Zinc Group** N=124
Who answered? (first respondent)		% (n)			
Child's mother		72.9 (638)	64.7 (216)	72.7 (56)	61.3 (76)
Child's father		4.0 (35)	6.9 (23)	6.5 (5)	7.3 (9)
Other relative (grandparent, aunt/uncle, sibling, other)		23.1 (202)	28.4 (95)	20.8 (16)	31.5 (39)
		% Yes (n)			
Q1: Motor milestone delay	Compared with other children, did your child have any serious delay in sitting, standing, or walking?	12.7 (111)	6.9 (23)	11.7 (9)	12.9 (16)
Q2: Vision	Compared with other children, does your child have difficulty seeing, either in daytime or nighttime?	1.1 (10)	0.9 (3)	0.0 (0)	0.0 (0)
Q3: Hearing	Does your child appear to have difficulty hearing?	1.7 (15)	2.1 (7)	1.3 (1)	4.8 (6)
Q4: Understanding	When you tell your child to do something, does s/he have trouble understanding what you are saying?	1.7 (15)	2.1 (7)	0.0 (0)	4.0 (5)
Q5: Movement	Does your child have difficulty walking or moving his/her arms or does s/he have weakness and/or stiffness in the arms or legs?	1.8 (16)	2.1 (7)	0.0 (0)	2.4 (3)
Q6: Fits/ seizures	Does your child have fits/convulsions, become rigid or lose consciousness?	1.0 (9)	2.4 (8)	0.0 (0)	3.2 (4)
Q7: Learning	Does the child have trouble learning to do things like other children of his/her age?	15.6 (136)	9.8 (32)	0.0 (0)	3.2 (4)
Q8: No speech	Is your child mute (note able to speak at all)?	2.9 (25)	3.0 (10)	1.3 (1)	1.6 (2)
Q9: Speech problems*	Is your child's speech in any way different from normal?	3.2 (20)	2.2 (5)	3.9 (3)	8.1 (10)
Q10: Slowness*	Compared with other children of his/her age, does your child appear in any way mentally slow, dull, or backward?	3.0 (19)	3.4 (8)	1.3 (1)	4.0 (5)
Q11: Behavioral problems*	Does your child show any behavioral problem, such as frequent tantrums, aggressive behavior, or difficulty relating to people?	3.2 (20)	5.6 (13)	3.9 (3)	4.0 (5)
Positive screen (yes to one or more questions)		31.1 (272)	24.3 (81)	20.8 (16)	31.5 (39)

Table 3 Results of Logistic Regression Analyses with TQP Outcomes of Interest on Iron/Zinc Supplementation Group, Intent-to-Treat Analysis, N=1410

Outcome **Treatment group**	**Positive** **Response n (%)**	**Crude OR** **(95% CI)**	**Adjusted* OR** **(95% CI)**
Positive Screen		†	
Placebo**	272 (31.1)	1.00	1.00
Zinc only	81 (24.3)	0.71 (0.53 - 0.95)	0.70 (0.51 - 0.94)
Iron only	16 (20.8)	*0.58 (0.33 - 1.03)*	0.71 (0.38 - 1.36)
Iron & zinc combination	39 (31.5)	1.02 (0.68 - 1.53)	1.23 (0.75 - 2.03)
Q1:Motor milestone delay		†	†
Placebo	111 (12.7)	1.00	1.00
Zinc only	23 (6.9)	0.51 (0.32 - 0.81)	0.57 (0.35 - 0.92)
Iron only	9 (11.7)	0.91 (0.44 - 1.87)	0.82 (0.36 - 1.87)
Iron & zinc combination	16 (12.9)	1.02 (0.58 - 1.78)	0.85 (0.43 - 1.67)
Q3:Hearing			
Placebo	15 (1.7)	1.00	1.00
Zinc only	7 (2.1)	1.23 (0.50 - 3.04)	1.34 (0.52 - 3.45)
Iron only	1 (1.3)	0.75 (0.10 - 5.78)	0.42 (0.05 - 3.73)
Iron & zinc combination	6 (4.8)	**2.91 (1.11 - 7.64)**	1.60 (0.47 - 5.49)
Q7: Learning		†	†
Placebo	136 (15.6)	1.00	1.00
Zinc only	32 (9.8)	0.59 (0.39 - 0.89)	0.56 (0.36 - 0.86)
Iron only	0 (0.0)	-	-
Iron & zinc combination	4 (3.2)	**0.18 (0.07 - 0.50)**	1.52 (0.41 - 5.62)
Q8: No speech			
Placebo	25 (2.9)	1.00	1.00
Zinc only	10 (3.0)	1.05 (0.50 - 2.22)	1.02 (0.47 - 2.21)
Iron only	1 (1.3)	0.45 (0.06 - 3.34)	0.74 (0.08 - 7.00)
Iron & zinc combination	2 (1.6)	0.56 (0.13 - 2.38)	1.02 (0.18 - 5.91)
Q9: Speech problems‡			

Outcome Treatment group	Positive Response n (%)	Crude OR (95% CI)	Adjusted* OR (95% CI)
Placebo	20 (3.2)	1.00	1.00
Zinc only	5 (2.2)	0.67 (0.25 - 1.81)	0.62 (0.22 - 1.71)
Iron only	3 (3.9)	1.24 (0.36 - 4.27)	0.81 (0.20 - 3.31)
Iron & zinc combination	10 (8.1)	**2.68 (1.22 - 5.88)**	1.81 (0.66 - 4.97)
Q10:Slowness‡			
Placebo	19 (3.0)	1.00	1.00
Zinc only	8 (3.4)	1.14 (0.49 - 2.63)	1.16 (0.48 - 2.83)
Iron only	1 (1.3)	0.42 (0.06 - 3.20)	0.22 (0.03 - 1.76)
Iron & zinc combination	5 (4.0)	1.35 (0.49 - 3.68)	0.71 (0.23 - 2.18)
Q11:Behavioral problems‡			
Placebo	20 (3.2)	1.00	1.00
Zinc only	13 (5.6)	1.80 (0.88 - 3.67)	1.75 (0.79 - 3.88)
Iron only	3 (3.9)	1.24 (0.36 - 4.27)	1.37 (0.33 - 5.64)
Iron & zinc combination	5 (4.0)	1.28 (0.47 - 3.49)	1.47 (0.44 - 4.88)

Bolded statistically significant with p-value < 0.05; *Italicized* 0.05 < p-value < 0.10

* Separate logistic regression models adjusted for child's sex and age of entry into study (yrs); family caste and land ownership (yes/no)

** Of N=1410: Placebo N=875 (62.1%), Zinc only N=334 (23.7%), Iron only N=77 (5.5%), Iron & zinc combination N=124 (8.8%)

† Likelihood ratio test p-value < 0.05

‡ Only asked of children 3 years or older (N=1067); Of N=1067: Placebo N=632 (59.2%), Zinc only N=234 (21.9%), Iron only N=77 (7.2%), Iron & zinc combination N=124 (11.6%)

As the number of iron tablets taken increased, the prevalence of a positive screen declined, then increased for those who took more than 350 tablets (Table 4). The prevalence of a positive screen declined as the number of zinc tablets taken increased (Table 4). The results of positive screening by number of iron or zinc tablets taken for the 3 most prevalent questions [motor

milestone delay (Q1), learning problems (Q7), and behavioral problems (Q11)] are also shown in Table 4.

Table 4 Percent Positive TQP Responses by Number of Tablets Taken, N=1410

Outcome	**Number of Iron Tablets Taken**				
	0	**1-60**	**>60-180**	**>180-350**	**>350-594**
N:	1209	50	52	50	49
Positive Screen	29.2	32.0	21.2	24.0	32.7
Q1: Motor milestones	11.1	14.0	13.5	12.0	10.2
Q7: Learning problems	14.0	2.0	3.9	0.0	2.0
N*:	865	50	50	50	51
Q11: Behavioral problems*	3.8	2.0	2.0	6.0	5.9
	Number of Zinc Tablets Taken				
	0	1-102	>102-266	>266-484	>484-1052
N:	952	116	115	114	113
Positive Screen	30.3	32.8	25.2	28.1	18.6
Q1: Motor milestones	12.6	10.3	7.0	8.9	8.0
Q7: Learning problems	14.3	15.2	10.5	5.4	0.9
N*:	708	91	90	90	87
Q11: Behavioral problems*	3.3	7.7	4.4	2.2	5.8

* Only asked of children 3 years or older (N=1067)

Analysis of the dose-response effect of number of iron and zinc tablets taken on positive TQP screening showed a statistically significant reduced odds for zinc (aOR=0.992, 95% CI: 0.986-0.999, for every 10 tablets taken) (Table 5). This effect was mainly driven by learning problems (aOR=0.972, 95% CI: 0.955-0.989). Because of the results in Table 4, we examined the iron effect using a spline model with cutoffs at 60 and 350 iron tablets. We did not find evidence of a differential effect of iron dose on risk of screening positive. Hence the linear model results are shown in Table 5. There was an increased prevalence of screen positivity for every 10 iron tablets taken (aOR=1.016, 95% CI: 1.003-1.029), being driven primarily by an increase among those who consumed less than 60 tablets of iron (see supplemental table). Though the odds ratios did not achieve statistical significance, there was evidence for increased report of motor milestone delay, hearing, learning, speech, learning, and behavioral problems with increasing iron tablet consumption.

Table 5 Results of Logistic Regression Analyses With TQP Outcomes of Interest on Number of Iron and Zinc Tablets Taken, N=1410

Outcome **Tablets taken (continuous)**	**Crude OR (95% CI)**	**Adjusted* OR (95% CI)**
Positive Screen		
Iron Tablets*	1.000 (0.989 - 1.012)	1.016 (1.003 - 1.029)†
Zinc Tablets*	0.991 (0.985 - 0.997)†	0.992 (0.986 - 0.999)†
Question 1:Motor milestone delay		
Iron Tablets*	1.004 (0.988 - 1.020)	1.005 (0.987 - 1.023)
Zinc Tablets*	0.989 (0.980 - 0.999)†	0.991 (0.981 - 1.001)
Question 3:Hearing		
Iron Tablets*	1.024 (0.997 - 1.051)	1.012 (0.982 - 1.043)
Zinc Tablets*	1.012 (0.999 - 1.026)	1.013 (0.998 - 1.027)†
Question 7: Learning		
Iron Tablets*	0.914 (0.861 - 0.969)†	1.032 (0.991 - 1.075)†
Zinc Tablets*	0.966 (0.952 - 0.981)†	0.972 (0.955 - 0.989)†
Question 8: No speech		
Iron Tablets*	0.975 (0.926 - 1.026)	1.007 (0.958 - 1.057)†
Zinc Tablets*	1.003 (0.989 - 1.018)	1.007 (0.992 - 1.022)†
Question 9: Speech problems‡		
Iron Tablets*	1.012 (0.987 - 1.038)	1.101 (0.896 - 1.353)
Zinc Tablets*	0.992 (0.975 - 1.008)	0.989 (0.972 - 1.007)
Question 10:Slowness ‡		
Iron Tablets*	0.995 (0.962 - 1.030)	0.987 (0.911 - 1.025)
Zinc Tablets*	1.001 (0.987 - 1.015)	1.001 (0.986 - 1.017)
Question 11:Behavioral problems‡		
Iron Tablets*	1.013 (0.990 - 1.037)	1.017 (0.990 - 1.044)
Zinc Tablets*	1.005 (0.992 - 1.017)	1.004 (0.992 - 1.017)

Bolded statistically significant with p-value < 0.05; *Italicized* 0.05 < p-value < 0.10

* Separate logistic regression model with number of tablets/10 rounded to the nearest 10 coded as an ordinal variable and adjusted for child's sex and age at TQP administration; family land ownership (yes/no) and caste

† Likelihood ratio test p-value < 0.05

‡ Only asked of children 3 years or older (N=1067)

Supplemental Table: Results of Logistic Regression Analyses With TQP Outcomes of Interest on Number of Iron Tablets Taken, N=1410

Outcome	0-60 Iron Tablets (n=1259)		61-350 Iron Tablets (n=102)		>350 Iron Tablets (n=49)	
	Crude OR (95% CI)	Adjusted* OR (95% CI)	Crude OR (95% CI)	Adjusted* OR (95% CI)	Crude OR (95% CI)	Adjusted* OR (95% CI)
Positive Screen	0.94 (0.77 – 1.15)	1.13 (0.92 – 1.38)	1.03 (0.98 – 1.09)	1.03 (0.96 – 1.10)	1.03 (0.95 – 1.13)	1.03 (0.93 – 1.14)
Q1:Motor milestone delay	1.06 (0.83 – 1.36)	1.06 (0.81 – 1.37)	1.02 (0.96 – 1.10)	1.01 (0.93 – 1.09)	1.12 (0.98 – 1.29)	1.22 (0.97 – 1.54)
Q3:Hearing	0.77 (0.26 – 2.29)	0.64 (0.20 – 2.09)	1.01 (0.89 – 1.16)	1.07 (0.86 – 1.34)	0.87 (0.68 – 1.12)	0.81 (0.56 – 1.17)
Q7: Learning	-	-	0.92 (0.75 – 1.12)	1.13 (0.73 – 1.74)	0.93 (0.66 – 1.31)	-
Q8: No speech	1.15 (0.79 – 1.68)	1.61 (1.07 – 2.44)†	-	-	1.07 (0.81 – 1.40)	0.93 (0.65 – 1.33)
Q9: Speech problems‡	0.97 (0.58 – 1.62)	0.88 (0.51 – 1.52)	0.89 (0.75 – 1.05)	0.97 (0.76 – 1.25)	0.88 (0.71 – 1.09)	0.82 (0.60 – 1.11)
Q10:Slowness ‡	-	-	0.95 (0.84 – 1.07)	-	0.39 (0.05 – 3.10)	-
Q11:Behavioral problems‡	0.94 (0.56 – 1.60)	0.96 (0.56 – 1.63)	1.06 (0.94 – 1.19)	1.14 (0.92 – 1.41)	1.08 (0.91 – 1.27)	1.06 (0.89 – 1.27)

Bolded statistically significant with p-value < 0.05; *Italicized* 0.05 < p-value ≤ 0.10

- Indicates empty cells and unable to run a logistic regression model

* Logistic regression model with number of iron tablets/10 rounded to the nearest 10 coded as an ordinal variable and adjusted for child's sex and age at TQP administration; family land ownership (yes/no) and caste

† Likelihood ratio test p-value < 0.05

‡ Only asked of children 3 years or older (N=1067; 0-60 iron tablets n=915; 61-350 iron tablets n=100; >350 tablets n=51; missing information for n=1)

Discussion

Roughly 30% of our sample screened positive for disability. Motor milestone delay and learning problems were the most prevalent reported problems. These results are consistent with TQ studies in other developing countries[27].

Our results indicate a reduced likelihood of TQP screen-positivity for childhood disability in the zinc only supplementation group. We found a similar effect when examining the number of zinc tablets taken, with decreased likelihood of screening positive as the number of tablets taken increased. Zinc supplementation was associated specifically with reduced likelihood of motor milestone and learning problems. Zinc supplementation promotes both physical and cognitive development, and studies have shown that zinc treatment can reduce recurrent episodes of diarrhea and pneumonia[28,29]. The effects we found could also be indirect in that zinc-replete children suffer less morbidity, which improves development[4].

There was no evidence of an effect of supplementation with iron onlyon screen positivity. Because the iron arms of the trial were halted roughly halfway through the study, there was a much smaller number of children analyzed in the iron only group compared to the other three groups, which reduced the power to detect any true effect. We also found a non-linear relationship with the dose of iron on screening positivity, with increased odds for 60 or fewer doses, but a null effect with higher doses. It is possible that the dose of iron given early in life was harmful. Given the well-documented association between iron deficiency and impaired cognitive development, the reasons for this result are not clear[30,31].

Children in the iron and zinc combination group also had no evidence for impact of supplements on screen positivity. A systematic review of trials in which children were given iron and zinc simultaneously have indicated decreased absorption of each micronutrient compared to when given separately. Whereas there appears to be no negative effect of zinc status with dual supplementation, iron indicators tend to be reduced[32]. This interactive effect of iron and zinc may explain the inconclusive results we found, although the biochemical absorption processes of micronutrients are not well understood[33].

Our study includes a population-based sample from rural south Asia large enough to detect odds ratios of public health importance for the prevalence of overall positive screening for the zinc versus placebo comparison. For detecting differences due to iron supplementation, however, sample size was a limiting factor. Compared to urban areas of Nepal, the study population had lower household wealth as indicated by ownership of common household items and a lower proportion of literate adult males, as might be expected[34]. However the study population was comparable to other rural areas of Nepal, and the results we found are likely applicable.

It should be noted that the TQP is a parental report of perceived disabilities or delay in development and does not represent a definitive diagnosis. However, because of the sensitivity of the TQP for diagnosis of developmental disabilities and delays, our results suggest that zinc supplementation may have an impact on learning problems and motor milestone delays, and that earlier age at initiation of supplementation and/or greater cumulative dose may increase this effect size. These findings support studies that have demonstrated an impact of early zinc supplementation on cognition and motor development.

Acknowledgements

We thank Dr. Naila Khan, head of the Child Development Centre, Child Development and Neurology Unit, Dhaka Shishu (Children's) Hospital, for her advice and training of our investigators in the administration of the TQP.

Funding

This study was conducted by the Department of International Health, Bloomberg School of Public Health, Johns Hopkins University (JHU), Baltimore Maryland, under grants from the National Institutes of Health, Bethesda, Maryland (HD38753), the Bill and Melinda Gates Foundation, Seattle, Washington (810-2054), the US Fund for UNICEF (50140) and a Cooperative Agreement between JHU and the Office of Health and Nutrition, US Agency for International Development, Washington DC (HRN-A-00-97-00015-00).

Disclosure

The authors have no financial relationships or competing interests relevant to this article to disclose. The corresponding and first authors had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

References

1. UNICEF. Global thematic report: integrated early child development, 2005.
2. UN. UN Convention on the Rights of People with Disabilities and Optional Protocol, 2006. Available at:http://www.un.org/disabilities/documents/convention/convoptprot-e.pdf.
3. Grantham-McGregor S, Cheung YB, Cueto S, Glewwe P, Richter L, Strupp B. Developmental potential in the first 5 years for children in developing countries. Lancet. 2007 Jan 6; 369(9555):60-70.
4. Walker SP, Wachs TD, Gardner JM, Lozoff B, Wasserman GA, Pollitt E, et al. Child development: risk factors for adverse outcomes in developing countries. Lancet. 2007 Jan 13;369(9556):145-57.
5. Black MM, Baqui AH, Zaman K, Ake Persson L, El Arifeen S, Le K, et al. Iron and zinc supplementation promote motor development and exploratory behavior among Bangladeshi infants. Am J ClinNutr. 2004 Oct;80(4):903-10.
6. Lozoff B, De Andraca I, Castillo M, Smith JB, Walter T, Pino P. Behavioral and developmental effects of preventing iron-deficiency anemia in healthy full-term infants. Pediatrics. 2003 Oct;112(4):846-54.
7. Bhutta ZA, Black RE, Brown KH, Gardner JM, Gore S, Hidayat A, et al. Prevention of diarrhea and pneumonia by zinc supplementation in children in developing countries: pooled analysis of randomized controlled trials. Zinc Investigators' Collaborative Group.J Pediatr. 1999 Dec;135(6):689-97.
8. Brown KH, Peerson JM, Rivera J, Allen LH. Effect of supplemental zinc on the growth and serum zinc concentrations of prepubertal children: a meta-analysis of randomized controlled trials. Am J ClinNutr. 2002 Jun;75(6):1062-71.
9. Sazawal S, Black RE, Menon VP, Dinghra P, Caulfield LE, Dhingra U, et al. Zinc supplementation in infants born small for gestational age reduces mortality: a prospective, randomized, controlled trial. Pediatrics. 2001 Dec;108(6):1280-6.

10. Bhutta ZA, Ahmed T, Black RE, Cousens S, Dewey K, Giugliani E, et al. What works? Interventions for maternal and child undernutrition and survival.Lancet. 2008 Feb 2;371(9610):417-40.
11. Bryce J, el Arifeen S, Pariyo G, Lanata C, Gwatkin D, Habicht JP. Reducing child mortality: can public health deliver? Lancet. 2003 Jul 12;362(9378):159-64.
12. Bryce J, Terreri N, Victora CG, Mason E, Daelmans B, Bhutta ZA, et al. Countdown to 2015: tracking intervention coverage for child survival. Lancet. 2006 Sep 23;368(9541):1067-76.
13. Jones G, Steketee RW, Black RE, Bhutta ZA, Morris SS. How many child deaths can we prevent this year? Lancet. 2003 Jul 5;362(9377):65-71.
14. Maulik PK, Darmstadt GL. Community-based interventions to optimize early childhood development in low resource settings.J Perinatol. 2009 Aug;29(8):531-42.
15. Stein Z, Belmont L, Durkin M. Mild mental retardation and severe mental retardation compared: experiences in eight less developed countries. Ups J Med Sci Suppl. 1987;44:89-96.
16. Thorburn M, Desai P, Paul TJ, Malcolm L, Durkin M, Davidson L. Identification of childhood disability in Jamaica: the ten question screen. Int J Rehabil Res. 1992;15(2):115-27.
17. UNICEF. Monitoring Child Disability in Developing Countries: Results from the Multiple Indicator Cluster Surveys (MICS) 2008.
18. Berg AL, Papri H, Ferdous S, Khan NZ, Durkin MS. Screening methods for childhood hearing impairment in rural Bangladesh. Int J PediatrOtorhinolaryngol. 2006 Jan;70(1):107-14.
19. Durkin MS, Davidson LL, Desai P, Hasan ZM, Khan N, Shrout PE, et al. Validity of the ten questions screened for childhood disability: results from population-based studies in Bangladesh, Jamaica, and Pakistan. Epidemiology. 1994 May;5(3):283-9.
20. Khan NZ, Ferdous S, Islam R, Sultana A, Durkin M, McConachie H. Behaviour problems in young children in rural Bangladesh. J Trop Pediatr. 2009 Jun;55(3):177-82.
21. Belmont L. Screening for severe mental retardation in developing countries: the International Pilot Study of Severe Childhood Disability. In: Berg J, editor. Science and technology in mental retardation. London: Methuen; 1986. p. 389-95.
22. Tielsch JM, Khatry SK, Stoltzfus RJ, Katz J, LeClerq SC, Adhikari R, et al. Effect of daily zinc supplementation on child mortality in southern Nepal: a community-based, cluster randomised, placebo-controlled trial. Lancet. 2007 Oct 6;370(9594):1230-9.
23. Tielsch JM, Khatry SK, Stoltzfus RJ, Katz J, LeClerq SC, Adhikari R, et al. Effect of routine prophylactic supplementation with iron and folic acid on preschool child mortality in southern Nepal: community-based, cluster-randomised, placebo-controlled trial. Lancet. 2006 Jan 14;367(9505):144-52.
24. Nepal living standards survey 2003/04, statistical report volume two. Kathmandu, 2004.
25. 25. SAS 9.2. Cary, NC. 2008.
26. StataCorp. Stata Statistical Software: Release 10. College Station, TX. 2007.
27. Gottlieb CA1, Maenner MJ, Cappa C, Durkin MS.Child disability screening, nutrition, and early learning in 18 countries with low and middle incomes: data from the third round of UNICEF's Multiple Indicator Cluster Survey (2005-06).Lancet. 2009 Nov 28; 374(9704):1831-9. doi: 10.1016/S0140-6736(09)61871-7.
28. Walker CL, Black RE. Zinc for the treatment of diarrhoea: effect on diarrhoea morbidity, mortality and incidence of future episodes. Int J Epidemiol. 2010 Apr;39Suppl 1:i63-9.
29. Mozaffari-Khosravi H, Shakiba M, Eftekhari MH, Fatehi F. Effects of zinc supplementation on physical growth in 2-5-year-old children. Biol Trace Elem Res. 2009 May;128(2):118-27.
30. Grantham-McGregor S, Ani C. A review of studies on the effect of iron deficiency on cognitive development in children.J Nutr. 2001;131(2S-2):649S-666S
31. Lozoff B, Beard J, Connor J, Barbara F, Georgieff M, Schallert T. Long-lasting neural and behavioral effects of iron deficiency in infancy. Nutr Rev. 2006;64(5 Pt 2):S34-43
32. Fischer Walker C, Kordas K, Stoltzfus RJ, Black RE. Interactive effects of iron and zinc on biochemical and functional outcomes in supplementation trials.Am J ClinNutr. 2005 Jul;82(1):5-12.
33. Sandstrom B. Micronutrient interactions: effects on absorption and bioavailability. Br J Nutr. 2001 May;85Suppl 2:S181-5.
34. Ministry of Health and Population (MOHP) [Nepal], New ERA, and Macro International Inc. 2007. Nepal Demographic and Health Survey 2006. Kathmandu, Nepal: Ministry of Health and Population, New ERA, and Macro International Inc.