Children's Intellectual Function in Relation to Arsenic Exposure

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Background: Very little evidence exists concerning the possible impairment of children's intellectual function in relation to arsenic exposure in utero and during childhood.

Methods: We conducted a cross-sectional study among 351 children age 5 to 15 years who were selected from a source population of 7683 people in West Bengal, India, in 2001–2003. Intellectual function was assessed with 6 subtests from the Wechsler Intelligence Scale for Children as well as with the Total Sentence Recall test, the Colored Progressive Matrices test, and a pegboard test. Arsenic in urine and lifetime water sources (including during the pregnancy period) were assessed using measurements of samples from 409 wells. The test scores were analyzed with linear regression analyses based on the method of generalized estimating equations incorporating relevant covariates.

Results: Stratifying urinary arsenic concentrations into tertiles, we found associations between arsenic and reductions in the adjusted scores of the vocabulary test (0, −0.14, −0.28; P for trend = 0.02), the object assembly test (0, −0.16, −0.24; P for trend = 0.03), and the picture completion test (0, −0.15, −0.26; P for trend = 0.02). These findings correspond to relative declines of 12% (95% confidence interval =0.4% to 24%) in the vocabulary test, 21% (−0.8% to 42%) in the object assembly test, and of 13% (0.3% to 24%) in the picture completion test in the upper urinary arsenic tertile. However, we did not find evidence of an association between test results and arsenic water concentrations during pregnancy or childhood.

Conclusions: Current arsenic concentrations in urine, which reflect all sources of recent exposure, including water and food, were associated with small decrements in intellectual testing in school-aged children in West Bengal. We did not see associations between long-term water arsenic concentrations and intellectual function.

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Children worldwide are exposed to arsenic in drinking water at concentrations that exceed the standard recommended by the World Health Organization and the U.S. Environmental Protection Agency maximum contaminant level of 10 μg/L.[1–3] Children are particularly at risk for high exposure in arsenic-affected areas of South Asia such as West Bengal, India.[4,5]

Acute neurotoxic effects of arsenic in high doses have been well documented.[6] Arsenic poisoning related to occupational exposure causes central nervous system alterations, including impairments of recent memory, learning, and concentration.[7,8] Children may be particularly susceptible to neurotoxic substances as suggested by findings from studies on the effects of lead,[9–11] methylmercury,[12,13] solvents,[14] and PCBs.[15] Experimental animal and in vitro studies,[16,17] and some limited evidence from the few earlier reports considering children's intellectual function and arsenic, suggested possible associations between arsenic exposure and neurodevelopment.[18–21] One recent study of 201 children conducted in Bangladesh, which is neighboring our study region, suggested that current arsenic concentrations in water as low as 10 μg/L were linked to reductions in intellectual functioning in 10-year-old children.[22]

We conducted a cross-sectional study of intellectual function among 351 children, aged 5 to 15 years, in the arsenic-exposed South-24-Parganas district in rural West Bengal, India, between 2001 and 2003.

METHODS

Study Location, Design, and Selection of Study Population

This cross-sectional study of children was conducted in 21 villages south of Kolkata in conjunction with a study of pregnancy outcomes.[23] Participants for the pregnancy study were selected based on the cross-sectional survey of 7683 people conducted in the same area in 1995–1996. The details of participant selection for the pregnancy outcome and the children studies are somewhat complex, because they were

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conducted in parallel with a study of chronic respiratory disease.

In the chronic respiratory disease study, a group of "high arsenic exposure" and a group of "low arsenic exposure" participants were identified from the previously mentioned cross-sectional survey. All participants from the survey who had drinking water arsenic concentrations greater than 400 μg/L and who also showed signs of arsenic-caused skin lesions were selected for the "high-exposure" group in the respiratory disease study. Participants in the "low-exposure" group of the respiratory disease study had drinking water containing less than 50 μg/L arsenic and had no signs of arsenic-caused skin lesions in 1995–1996. For each "high-exposure" participant, a "low-exposure" participant was randomly selected from those who matched by age (within 5 years) and sex. If a participant in the respiratory disease study was a married woman age 20 to 40 years, she was invited to participate in the pregnancy study. If not, then married women in the same household as the respiratory disease study participant, or close female neighbors drinking the same water in 1995–1996, and currently age 20 to 40 years, were invited to participate. A total of 205 women matching the selection criteria were identified and approached, and 203 agreed to participate (99%). The child study consisted of the participating women's children who were age 5 to 15 years at the time the study was conducted (n = 351). The participation among eligible children was 100%.

Informed consent was obtained from the parents of all children. The study was approved by the Institutional Review Boards of the Institute of Postgraduate Medical Research and Education, Kolkata, India, and the University of California, Berkeley, California.

Interview and House Visits

The field team, including a physician and a child psychologist, visited each family and arranged an appointment in the afternoon of a subsequent day after the child returned from school. During this second visit, the mother of the child was interviewed concerning sociodemographic factors, including parental education and occupation, location of the child's school (current and past), and lifetime residential history. The interviewer recorded the construction material of the house (mud, brick) and its number of rooms as a measure of socioeconomic status. After the intellectual function testing was completed, the field worker estimated the child's average daily water consumption using the container the child usually used for drinking to assess the volume of water consumed per day.

Children's Intellectual Function Testing

There was no intelligence test specifically designed for Bengali children. We therefore selected a series of tests and subtests familiar to local investigators that have been used under comparable conditions and that were thought to be culturally appropriate and could work across differences in language and lifestyle in rural West Bengal.[22,24,25] From the Wechsler Intelligence Scale for Children, we selected 2 of the verbal tests (vocabulary, digit span) and 4 of the performance-type subtests (object assembly, coding, picture completion, block design).[26,27] In addition, the Raven Colored Progressive Matrices test,[28] the Total Sentence Recall test,[29] and a version of the Purdue pegboard test suitable for children from the age of 5 years were administered. In the Total Sentence Recall test, sentences of increasing difficulty were administered to the following age groups: 5 to 6 years, 7 to 10 years, 11 to 12 years, and 13 to 15 years; these were scored for errors (omissions, additions, substitutions) with higher scores indicating more errors. For the pegboard test, the Colored Progressive Matrices test, and the Total Sentence Recall test, children age 5 to 15 years were included. For all other tests, children were aged 6 to 15 years. The same child psychologist administered all tests without knowledge of the child's arsenic exposure.

Physical Examination for Skin Lesions and Urine Samples

At the beginning of the visit, a soft drink was given to the child. At the end of the assessment, the height and weight of the child were measured, and the child was asked to provide a urine sample. The field physician carefully examined the child for possible arsenic-induced skin lesions in a well-lit area outdoors under natural light. Urine samples were transported on dry ice to the laboratory in Kolkata. They were stored at −20°C until analyses for total inorganic arsenic by flow injection analysis using atomic fluorescence detection with in-line photooxidation and continuous hydride generation.[30] The lower limit of quantification was <1 μg/L.

Arsenic in Water Exposure

The field team collected water samples from all functioning tube wells used by participants for at least 6 months during their lifetimes, including wells in schools. Some wells had been closed due to damaged filters or other mechanical problems or because they were known to have arsenic contamination. For some of these closed tube wells, we obtained past arsenic water concentrations measured before they were closed (n = 48). We collected samples of 361 functioning tube wells in the 21 villages in the study region. Private tube wells were often used by just one household, whereas government tube wells were used by multiple families. Water samples were stored in a cooler containing an ice block and transported to the laboratory in Kolkata on the same day. The water samples were then kept frozen at −20°C until they were analyzed. Total water arsenic was measured by flow injection analysis using atomic fluorescence detection with in-line photooxidation and continuous hydride generation.[30] The lower limit of quantification was <1 μg/L.

Statistical Analyses

Children's arsenic lifetime exposure histories were based on information about all tube wells used for at least 6 months at each residence and school and on the results of the arsenic measurements. Some children had at times used surface pond water for drinking. Because the arsenic concentrations were very low or nondetectable in initial pond test samples (range, <1–4.2 μg/L), we used zero as the concentration for all subsequent pond water sources. Children's annual average arsenic water concentrations were first calculated for every calendar year based on the measured water concentrations for each tube well used in that year and the fraction of drinking water children obtained from the respective source in that year (eg, 75% home well, 25% school well). Peak exposure (μg/L) was then defined as the highest known annual average water concentration of arsenic ingested by a child. Prenatal arsenic exposure for each child was assessed based on the mother's exposure during the months of pregnancy. Arsenic values were missing for 9 children for a certain time period since birth; the mean time period missing was 4.0 years, and these years were set to zero. For 12 children, arsenic values were missing during a portion of the pregnancy period (mean time period missing, 7.2 months) and the known arsenic value was applied to the entire pregnancy. Seven children had no arsenic measurement available for the pregnancy period and were excluded from the analyses in respect to arsenic during pregnancy.

We first performed univariate analyses to assess the children's sociodemographic factors, body mass index (BMI), and other relevant basic characteristics, the arsenic concentrations in water and urine, and the raw intellectual function test scores. Raw test scores for the Wechsler scale were used in the absence of norms for the Indian population. Descriptive age-stratified analyses were conducted to evaluate arsenic effects separately for each age comparing mean scores in different exposure categories using *t* tests. The associations between the intellectual function test scores and concentrations of arsenic were further assessed in linear regression models based on the method of generalized estimating equations (GEE) accounting for multiple children from the same mother.[31] The regression coefficients were standardized by dividing them by the standard deviation (SD) of each test so that they could be compared one with another.

Covariates were evaluated in univariate analyses and then in addition to arsenic in the regression models. The multivariate models included age (indicator variable for each year of age), sex, BMI, maternal and paternal education (no formal education, primary, secondary, and higher), father's occupation (unemployed, farming, daily wage, service vs business), mother's age, type of house building material (mud, brick, mixed material), and number of rooms in the house.

Arsenic concentrations in water (lifetime peak, lifetime cumulative, during pregnancy) and in urine were considered separately as continuous and categorical variables. Spearman correlation coefficients were computed to assess the correlation between the different arsenic exposure measures. Water concentrations were stratified in categories of 0–9 μg/L, 10–49 μg/L, 50–99 μg/L, and $\geq$100 μg/L, and urine concentrations were used as tertiles (cut points: Colored Progressive Matrices, Total Sentence Recall, and pegboard tests: 44.2 μg/L, 86.1 μg/L; all other tests: 43.6 μg/L, 82.6 μg/L). For the overall evaluation of children's intellectual function, we added z-scores of the 2 selected verbal tests and 4 selected performance type subtests of the Wechsler scale to obtain a "Full-Scale" summary measure.[27] Like with the individual tests, the regression coefficients of the summary measure were divided by the SD of the summary measure. Tests for trends across exposure categories were computed by forming a variable with the increasing levels of arsenic (coded 0, 1, 2, for urine; 0, 1, 2, 3 for water) and by including this variable in the respective linear GEE models adjusted for the same covariates.[32] Because we had a clear a priori hypotheses, we calculated one-sided *P* values for trends. All data analyses were carried out using the SAS statistical program package (version 8.0e; SAS Institute, Cary, NC).

RESULTS

Median age was 9 years, and 46% of the sample were girls (Table 1). We obtained complete exposure histories for 342 (97%) of the children for the time since birth and for 332 (95%) children during pregnancy. The average lifetime peak arsenic concentration was 147 μg/L with a maximum of 2480 μg/L (Table 2). Urine samples were provided by 99% (n = 349)

TABLE 1. Baseline Characteristics of the Children in West Bengal (n = 351)

Characteristic	
Age distribution in years; %	
5–7	30
8–11	42
12–15	29
Mother's age (years); median (range)	31 (20–41)
BMI (kg/m^2); median (range)	14.1 (10.5–32.3)
Sex; %	
Girls	46
Boys	54
Maternal education; %	
No formal	39
Primary	27
Secondary and higher	33
Paternal education; %	
No formal	24
Primary	29
Secondary and higher	46
Unknown	1
Paternal occupation; %	
Unemployed	4
Farming	33
Business	22
Daily wage	23
Service	19
Maternal occupation; %	
Housekeeping	86
Daily wage	8
Other	6
Type of house; %	
Mud	37
Mixed	40
Brick	23
Rooms in house; %	
1	55
>1	44
Missing	1

TABLE 2. Children's Arsenic Exposure in Drinking Water and in Urine

Arsenic Exposure Measure	Mean ± SD (μg/L)	(Range)
Peak lifetime water*	147 ± 322	(1–2480)
Average lifetime water*	59 ± 133	(1–870)
Pregnancy water†	110 ± 243	(1–2536)
Current child urine‡	78 ± 61	(2–375)

*Based on annual average for all water sources used for at least 6 mo.
†Excludes 7 pregnancies with no exposure information.
‡Available for n = 349 (99%).

of the children. Arsenic concentrations in urine and peak water were slightly higher in boys (urine, mean ± SD = 87 ± 63 μg/L; water, 157 ± 355 μg/L) than in girls (68 ± 56 μg/L, $P = 0.005$; 136 ± 279 μg/L, $P = 0.5$). Peak lifetime arsenic exposure in water was 0–9 μg/L for 56% of children, 10–49 μg/L for 10%, 50–99 μg/L for 6%, and ≥100 μg/L for 29%.

A matrix of Spearman correlation coefficients among water and urine measurements is presented in Table 3. Strong correlations were observed between peak and lifetime arsenic exposure (r = 0.92). On average, girls consumed 1.5 L and boys 1.7 L water per day and the children had used 4 (range, 1–10) water sources during their lifetimes. No child was found to have arsenic-related skin lesions.

Sociodemographic variables included in the adjusted GEE-based regression models showed the expected associations with the children's test scores. In general, the strongest effects were seen for maternal education with children of mothers having "primary" and "secondary" education performing better than children of mothers with no formal education. For example, in the vocabulary test regression analysis, the coefficient for maternal primary education was 0.23 (95% confidence interval [CI] = −0.05 to 0.50) and 0.47 (0.17 to 0.76) for maternal secondary education. Effects of paternal education and other sociodemographic variables were weaker than the effects of maternal education. Having more than one room in the house was also related to better test results such as in the vocabulary test (0.2; −0.08 to 0.4).

No clear pattern was found for increasing categories of peak arsenic water concentrations since birth and children's scores in the various tests (Table 4). Although the adjusted coefficients in the vocabulary, coding and Colored Progressive Matrices tests were reduced in the exposure categories above 10 μg/L as compared with those for arsenic concen-

TABLE 3. Association Among Measures of Arsenic Exposure in Drinking Water and Urine (Spearman Correlation Coefficients)

	Lifetime Average	Pregnancy	Urinary
Peak	0.92	0.69	0.11
Lifetime average		0.71	0.12
Pregnancy			0.07

TABLE 4. Children's* Unadjusted and Adjusted† Intellectual Function Test Results (95% CIs) and Peak Arsenic in Drinking Water Concentrations in Categories and as Continuous Variable (adjusted†) in GEE-Based Linear Regression Models‡

Test	10–49 μg/L Unadjusted	10–49 μg/L Adjusted†	50–99 Arsenic (μg/L) Unadjusted	50–99 Arsenic (μg/L) Adjusted†	≥100 μg/L Unadjusted	≥100 μg/L Adjusted†	P for Trend†§	Continuous† per 100 μg/L
Vocabulary	−0.36 (−0.67 to −0.05)	−0.17 (−0.48 to 0.14)	−0.38 (−0.76 to 0.005)	−0.23 (−0.59 to 0.12)	0.18 (−0.09 to 0.45)	−0.05 (−0.29 to 0.20)	0.3	0.01 (−0.02 to 0.04)
Digit span	−0.16 (−0.54 to 0.22)	0.08 (−0.24 to 0.40)	−0.26 (−0.78 to 0.26)	−0.15 (−0.54 to 0.23)	0.12 (−0.17 to 0.40)	−0.08 (−0.32 to 0.17)	0.2	0.02 (−0.01 to 0.05)
Object assembly	−0.005 (−0.44 to 0.43)	0.16 (−0.23 to 0.55)	−0.08 (−0.51 to 0.35)	0.014 (−0.43 to 0.46)	0.28 (−0.006 to 0.57)	0.06 (−0.18 to 0.31)	0.7	0.02 (−0.02 to 0.06)
Coding	−0.34 (−0.70 to 0.02)	−0.14 (−0.47 to 0.20)	−0.20 (−0.84 to 0.44)	−0.03 (−0.48 to 0.43)	0.054 (−0.22 to 0.33)	−0.13 (−0.37 to 0.11)	0.2	0.01 (−0.02 to 0.04)
Picture completion	−0.10 (−0.43 to 0.23)	0.12 (−0.19 to 0.43)	−0.63 (−1.10 to −0.17)	−0.45 (−0.84 to −0.07)	0.13 (−0.15 to 0.41)	−0.09 (−0.33 to 0.14)	0.1	0 (−0.03 to 0.04)
Block design	−0.16 (−0.43 to 0.11)	−0.01 (−0.25 to 0.23)	0.004 (−0.50 to 0.51)	0.05 (−0.33 to 0.44)	0.18 (−0.9 to 0.46)	−0.02 (−0.23 to 0.22)	0.5	0.02 (−0.02 to 0.05)
Pegboard	−0.23 (−0.64 to 0.18)	−0.10 (−0.39 to 0.19)	0.08 (−0.45 to 0.61)	0.13 (−0.27 to 0.53)	0.33 (0.10 to 0.57)	0.06 (−0.14 to 0.26)	0.7	0.01 (−0.02 to 0.003)
CPM	−0.21 (−0.52 to 0.10)	−0.02 (−0.28 to 0.24)	−0.31 (−0.66 to 0.05)	−0.29 (−0.57 to −0.02)	0.27 (−0.007 to 0.54)	−0.02 (−0.28 to 0.24)	0.4	0.01 (−0.02 to 0.04)
TSR	0.10 (−0.32 to 0.51)	0.11 (−0.19 to 0.41)	0.42 (−0.06 to 0.90)	0.28 (−0.08 to 0.64)	0.009 (−0.24 to 0.26)	−0.03 (−0.27 to 0.21)	0.5	−0.03 (−0.05 to 0)
Full Scale	−0.21 (−0.57 to 0.15)	0.006 (−0.31 to 0.33)	−0.32 (−0.83 to 0.20)	−0.16 (−0.56 to 0.23)	0.19 (−0.10 to 0.49)	−0.06 (−0.30 to 0.18)	0.3	−0.02 (−0.02 to 0.05)

*Age groups: Pegboard, CPM, and TSR: 5–15 years (n = 351), all others 6–15 years (n = 326).
†Adjusted for age using indicator variables for each age, sex, maternal and paternal education (no formal education, primary, secondary, and higher), father's occupation (unemployed, farming, daily wage, service, business), number of rooms in the house, type of house building material (mud, brick, mixed material), BMI, and mother's age.
‡Beta coefficients divided by the standard deviation of each test.
§Test for trends across categories, adjusted analysis, one-sided.
CPM indicates Colored Progressive Matrices; TSR, Total Sentence Recall.

trations below 10 μg/L, no dose–response associations were apparent. None of the other tests showed a dose–response relation. Using peak arsenic as a continuous variable in the regression models also did not support an adverse effect on the test results (Table 4). In the combined Full-Scale measure, the largest reduction was found in the intermediate category (−0.16; 95% CI = −0.56 to 0.23). Likewise, in utero arsenic exposure did not suggest an association with the test scores (Table 5). Using lifetime average or cumulative arsenic concentrations as exposure variables resulted in similar findings (data not shown).

Increasing tertiles of urinary arsenic concentrations were associated with reductions in some of the children's test scores (Table 6). The strongest effects were seen in the vocabulary test (2nd tertile, −0.14 [95% CI = −0.37 to 0.10]; 3rd tertile, −0.28 [−0.55 to −0.008]; *P* for trend = 0.02), the object assembly test (2nd tertile, −0.16 [−0.34 to 0.06]; 3rd tertile, −0.24 [−0.49 to 0.01]; *P* for trend = 0.03), and the picture completion test (2nd tertile, −0.15 [−0.34 to 0.09]; 3rd tertile, −0.26 [−0.51 to −0.01]; *P* for trend = 0.02) (Table 6). The stratum-specific estimates of all tests were inversely related to urinary arsenic except for the peg board test, and the lower categories of the block design and Colored Progressive Matrices tests. The adjusted Full-Scale summary of z-scores was reduced in a dose–response manner in association with increasing tertiles of urinary arsenic (*P* for trend = 0.05). However, when urinary arsenic concentrations were incorporated as a continuous variable, the associations were weaker than found with stratification in tertiles (right column of Table 6).

The effects seen in the upper tertile of urinary arsenic for the vocabulary, the object assembly, and the picture completion tests correspond to a reduction in the means of children's test scores of 12% (95% CI = 0.4% to 24%), 21% (−0.8% to 42%), and 13% (0.3% to 24%), respectively.

DISCUSSION

This study systematically addresses arsenic exposure from all water sources used over a lifetime (including the pregnancy period), as well as urinary arsenic concentrations, in relation to intellectual function in children. Effects were found for the vocabulary, picture completion, and object assembly tests with reductions between 12% and 20%, but the confidence intervals were broad. Our findings suggest that increased urinary arsenic concentrations reflecting current exposure from all sources, including food, are associated with small decrements in intellectual function testing, whereas little evidence for an effect of long-term arsenic concentrations in drinking water was found.

Limited data are available on the association between arsenic and intellectual function in children. In Thailand, arsenic concentrations in the hair of 529 children age 6 to 9 years were unrelated to test scores measured by a Motor-Free Visual Perception test and a Visual Motor integration test in the various arsenic-in-hair exposure groups.[19] Comparing high school students from an arsenic-affected area with a nonaffected area in Taiwan (n = 109), poorer results were found in 3 of 4 tests in children from the exposed area. However, no individual arsenic measurements were avail-

TABLE 5. Children's* Unadjusted and Adjusted† Intellectual Function Test Results (95% CIs) and Average Pregnancy Arsenic in Drinking Water Concentrations in Categories and as Continuous Variable (Adjusted†) in GEE-Based Linear Regression Models‡

Test	10–49 μg/L		50–99 Arsenic (μg/L)		≥100 μg/L		*P* for Trend†§	Continuous† per 100 μg/L
	Unadjusted	Adjusted†	Unadjusted	Adjusted†	Unadjusted	Adjusted†		
Vocabulary	−0.43 (−0.74 to −0.13)	−0.23 (−0.54 to 0.08)	−0.14 (−0.57 to 0.29)	−0.036 (−0.40 to 0.33)	0.06 (−0.23 to 0.34)	−0.08 (−0.26 to 0.53)	0.2	0.01 (−0.03 to 0.06)
Digit span	−0.21 (−0.61 to 0.18)	0.08 (−0.24 to 0.40)	−0.004 (−0.61 to 0.60)	0.09 (−0.36 to 0.54)	0.025 (−0.27 to 0.32)	−0.06 (−0.30 to 0.19)	0.4	0.01 (−0.02 to 0.04)
Object assembly	−0.086 (−0.51 to 0.34)	0.079 (−0.31 to 0.47)	0.049 (−0.35 to 0.45)	0.12 (−0.28 to 0.51)	0.28 (−0.02 to 0.58)	0.17 (−0.09 to 0.42)	0.9	0.02 (−0.01 to 0.06)
Coding	−0.32 (−0.69 to 0.05)	−0.13 (−0.48 to 0.21)	−0.21 (−0.81 to 0.39)	−0.08 (−0.51 to 0.36)	0.16 (−0.14 to 0.45)	0.031 (−0.21 to 0.27)	0.5	0.01 (−0.03 to 0.04)
Picture completion	−0.16 (−0.49 to 0.17)	0.08 (−0.24 to 0.40)	−0.40 (−0.84 to 0.05)	−0.25 (−0.58 to 0.09)	0.06 (−0.22 to 0.35)	−0.06 (−0.29 to 0.17)	0.2	−0.01 (−0.04 to 0.02)
Block design	−0.23 (−0.50 to 0.03)	−0.09 (−0.34 to 0.17)	0.11 (−0.35 to 0.56)	0.12 (−0.25 to 0.49)	0.10 (−0.19 to 0.39)	−0.01 (−0.26 to 0.23)	0.5	−0.02 (−0.05 to 0.02)
Pegboard	−0.33 (−0.68 to 0.028)	−0.18 (−0.44 to 0.09)	0.094 (−0.36 to 0.54)	0.09 (−0.29 to 0.48)	0.13 (−0.12 to 0.39)	−0.03 (−0.23 to 0.17)	0.5	0 (−0.02 to 0.03)
CPM	−0.29 (−0.62 to 0.036)	−0.08 (−0.36 to 0.20)	−0.066 (−0.46 to 0.32)	−0.07 (−0.38 to 0.24)	0.14 (−0.13 to 0.41)	−0.07 (−0.30 to 0.17)	0.3	0 (−0.03 to 0.03)
TSR	−0.02 (−0.39 to 0.35)	0.03 (−0.24 to 0.31)	0.43 (−0.006 to 0.87)	0.32 (−0.04 to 0.69)	−0.04 (−0.33 to 0.24)	−0.05 (−0.30 to 0.19)	0.6	−0.03 (−0.07 to 0.01)
Full Scale	−0.28 (−0.64 to 0.09)	−0.047 (−0.38 to 0.28)	−0.12 (−0.62 to 0.37)	−0.007 (−0.36 to 0.34)	0.14 (−0.17 to 0.45)	−0.002 (−0.24 to 0.24)	0.4	0.01 (−0.02 to 0.03)

*Children with known arsenic concentration in pregnancy, n = 344. Age groups: Pegboard, CPM, and TSR: 5–15 years (n = 344), all others 6–15 years (n = 319).
†Adjusted for same variable as in Table 4.
‡Beta coefficients divided by the standard deviation of each test.
§Test for trends across categories, adjusted analysis, one-sided.

TABLE 6. Children's* Unadjusted and Adjusted† Intellectual Function Test Results (95% CIs) and Urinary Concentrations of Arsenic in the Second and Third Tertile‡ vs the Lowest Tertile and as Continuous Variable (Adjusted†) in the GEE-Based Linear Regression Models¶

	2nd Tertile		3rd Tertile			
Test	Unadjusted	Adjusted†	Unadjusted	Adjusted†	P Trend†§	Continuous† per 100 μg/L
Vocabulary	−0.20 (−0.47 to 0.06)	−0.14 (−0.37 to 0.10)	−0.27 (−0.53 to −0.007)	−0.28 (−0.55 to −0.008)	0.02	−0.09 (−0.3 to 0.07)
Digit span	−0.15 (−0.42 to 0.13)	−0.04 (−0.30 to 0.22)	−0.05 (−0.32 to 0.22)	−0.0004 (−0.27 to 0.27)	0.5	0.04 (−0.1 to 0.2)
Object assembly	−0.16 (−0.43 to 0.11)	−0.16 (−0.34 to 0.06)	−0.15 (−0.43 to 0.12)	−0.24 (−0.49 to 0.01)	0.03	−0.07 (−0.2 to 0.1)
Coding	−0.27 (−0.53 to −0.005)	−0.14 (−0.37 to 0.10)	−0.16 (−0.43 to 0.11)	−0.13 (−0.38 to 0.12)	0.2	−0.06 (−0.2 to 0.09)
Picture completion	−0.21 (−0.5 to 0.07)	−0.15 (−0.34 to 0.09)	−0.27 (−0.53 to −0.003)	−0.26 (−0.51 to −0.01)	0.02	−0.10 (−0.3 to 0.04)
Block design	0.018 (−0.25 to 0.29)	0.076 (−0.16 to 0.31)	−0.041 (−0.31 to 0.23)	−0.085 (−0.34 to 0.17)	0.3	−0.02 (−0.2 to 0.2)
Pegboard	0.11 (−0.13 to 0.36)	0.15 (−0.07 to 0.36)	0.07 (−0.19 to 0.33)	0.09 (−0.14 to 0.32)	0.2	0.04 (−0.1 to 0.2)
CPM	−0.073 (−0.34 to 0.19)	0.0009 (−0.22 to 0.23)	−0.15 (−0.42 to 0.12)	−0.12 (−0.36 to 0.11)	0.2	−0.07 (−0.2 to 0.07)
TSR	0.34 (0.09 to 0.60)	0.23 (0.02 to 0.44)	0.14 (−0.11 to 0.39)	0.13 (−0.09 to 0.35)	0.9	0.04 (−0.1 to 0.2)
Full Scale	−0.20 (−0.47 to 0.08)	−0.11 (−0.34 to 0.12)	−0.19 (−0.44 to 0.07)	−0.20 (−0.44 to 0.03)	0.05	−0.07 (−0.2 to 0.09)

*Children with urinary arsenic measurements, n = 349. Age groups: Pegboard, CPM, and TSR: 5–15 years (n = 349), all others 6–15 years (n = 325).
†Adjusted for same variables as in Table 4.
‡Cutoff points: Pegboard, CPM, and TSR: 44.2 μg/L, 86.1 μg/L; all others: 43.6 μg/L, 82.6 μg/L.
§Test for trends across tertiles in the adjusted analysis, one-sided.
¶Beta coefficients divided by the standard deviation of each test.

able.[20] In Mexican children living close to a smelter (n = 41), increasing levels of urinary arsenic correlated with decreasing scores in verbal intelligence compared with children from a reference area (n = 39).[18] In a recent study from Bangladesh of 201 children (age 10 years), the authors reported a significant decrease in Full-Scale raw scores of 6.4 points in those currently drinking water containing arsenic above 50 μg/L as compared with a reference group with arsenic <5.5 μg/L.[22] A somewhat smaller effect on the Full-Scale raw scores was reported with urinary arsenic (−2.9 points). The magnitudes of the reductions in the Full-Scale scores found in Bangladesh are comparable to our findings with urinary arsenic, although we did not see the same association using different lifetime measurements of arsenic in drinking water. In our study, verbal and performance type testing were both affected, like in the study in Bangladesh.[22]

Exposure to arsenic during pregnancy was not associated with reduced scores in school-age in our study (Table 5). No previous investigation has addressed pregnancy exposures in relation to later intellectual testing, and this finding needs further confirmation. Children's lifetime consumption of arsenic in drinking water, including the pregnancy period, was assessed carefully in our study. On average, a 45-minute interview was conducted with the family to ascertain all water sources (including schools) followed by collection of water samples from accessible wells. Like in most environmental epidemiologic studies, there is uncertainty in past exposure assessment. However, with the same kind of lifetime exposure assessment, we have shown strong effects of arsenic in drinking water in the same study region on several outcomes, including skin lesions,[33] reduced lung function,[34] and increased risk for bronchiectasis[35] and stillbirths.[23]

Average urinary arsenic concentrations were relatively high with a mean of 78 μg/L and the 25th percentile at 110 μg/L. This indicates that children are still being exposed although their drinking water concentrations are lower than in the past. The correlation between arsenic concentrations in urine and in current drinking water was weak (r = 0.12). This may suggest that children are additionally exposed to arsenic from their diet (which is usually produced locally) where arsenic-contaminated water is also used for irrigation. Recent investigations on arsenic concentrations in food from affected areas in West Bengal and Bangladesh have shown contamination of locally grown food stuff, which comprises the typical diet of the children in our study area, ie, vegetables (range, 70–3990 μg/kg[36]; 20.9–21.2 μg/kg[37]; 5–540 μg/L[38]), cereals and bakery goods (mean, 130 μg/kg and 179 μg/kg[37]; 160 μg/kg[38]), rice (mean, 496 μg/kg; range, 58–1830 μg/kg[39]), spices (range, 133–202 μg/kg[37]), and freshwater fish (range, 97–1318 μg/kg[38]). These findings suggest that, in addition to drinking water, food could be an important source of children's arsenic exposure. Water concentrations on their own may not give a complete assessment of children's exposure.

We chose testing methods that have been applied previously in comparable study populations and conditions to assess intellectual child development and that were found to provide meaningful results.[24,25,40,41] All tests in our study were selected in consultation with the University of Kolkata and conducted by one local child psychologist in the Bengali language. The child psychologist was specifically trained in the field testing procedures and was unaware of children's exposure status. The underlying rationale of test selection was to measure verbal and performance abilities in children to identify reductions in certain abilities in relation to exposure. However, these tests have been standardized for the U.S. population and have inherent limitations when given to children from very different cultural backgrounds and levels of socioeconomic development. Thus, instead of using the U.S. norms, and in the absence of Indian norm values, we used children's "raw" scores in the multivariate regression analyses to assess effects of arsenic while adjusting for relevant covariates.

Our findings with urinary arsenic were stronger in the stratified analyses as compared with analyses that included urinary arsenic as a continuous variable. The findings with urinary arsenic as a continuous variable are strongly influenced by a small number of participants with very high urinary arsenic concentrations. The greater statistical power normally achieved with using continuous variables depends on linearity in the dose–response relationship. It is possible that effects occur in the highest stratum of urinary arsenic concentrations but without much further impact for the most highly exposed.

Careful attention was given to age adjustment. Indicator variables were included in the regression models for each year of age of children, thus accounting for any possible nonlinear relationships between age and intellectual testing. Socioeconomic status and parental education are commonly known to be associated with children's intellectual abilities, and we found strong effects of maternal education on test results. Somewhat weaker effects—but in the same expected direction—were seen for paternal education and the other sociodemographic variables such as the house's building material and number of rooms (considered good indicators of the socioeconomic living conditions of a family). Although socioeconomic variables were not related to children's arsenic exposure, we included a set of socioeconomic indicators, and maternal and paternal education and occupation variables in our analyses.

Experimental in vitro and in vivo studies provide biologic evidence of the central nervous system toxicity of arsenic. Most investigations considered short-term effects. In rodents exposed to very high doses of arsenic, the most common change in behavior reported was decreased locomotor activity.[16,17] Various neurotransmitters have been suspected of being involved in the mechanism of arsenic neurotoxicity, but the results are conflicting.[16] Oxidative stress reactions may be involved as arsenite inhibits glutathione reductase in brain tissue.[42] However, in vivo inhibition of glutathione reductase was only found at very high concentrations of arsenic.[43] Although in the latter study, arsenic metabolites MMA and DMA were measured in brain tissue of mice, the mechanism by which arsenic crosses the blood–brain barrier and the role of arsenic methylation in neurotoxicologic effects are not known.

Our findings suggest that arsenic exposure measured in urine is related to decrements in intellectual function scores, which may be in the range of 10% to 20% for some tests. However, the 95% confidence intervals of these estimates were wide. Whether or not these effects have persisting impact needs further investigation.[44] There was little evidence of an association between arsenic drinking water concentrations alone and intellectual function. Current urine concentrations reflecting exposure from all sources appeared to be more relevant than pregnancy, peak, or cumulative exposure based on measurements of water sources. One possible explanation is that the relationship with current exposure relates only to transient effects. However, it is also possible that the lack of findings with past water concentrations is due to incomplete assessment of past exposure, in particular, exposure originating from food. Although the findings need to be confirmed, they add to the body of evidence of adverse health effects in children resulting from exposure to arsenic.

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