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The association of urine markers of iodine intake with development and growth among children in rural Uganda: a secondary analysis of a randomised education trial

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Abstract

Objective: We examined associations of urine iodide excretion, proxy for iodine intake, with child development and growth.

Design: This is a secondary analysis of a 1:1 cluster-randomised trial with a 6-month nutrition/stimulation/hygiene education intervention among mothers of children aged 6–8 months to improve child development and growth. Development was assessed using Bayley Scales of Infant and Toddler Development–III (BSID-III) and Ages and Stages Questionnaire (ASQ), whereas anthropometry was used to assess growth. Urine iodide concentration (UIC) and urine iodide/creatinine ratio (ICR) were measured.

Setting: The current study was conducted in southern Uganda.

Participants: We randomly selected 155 children from the 511 enrolled into the original trial and analysed data when they were aged 20–24 and 36 months.

Results: Median UIC for both study groups at 20–24 and 36 months were similar ($P > 0{\cdot}05$) and within the normal range of 100–199 μg/l (0·79–1·60 μmol/l), whereas the intervention group had significantly higher ICR at 20–24 months. The BSID-III cognitive score was positively associated ($P = 0{\cdot}028$) with ICR at 20–24 months in the intervention group. The ASQ gross motor score was negatively associated ($P = 0{\cdot}020$) with ICR at 20–24 months among the controls. ICR was not significantly associated with anthropometry in the two study groups at either time-point.

Conclusions: Following the intervention, a positive association was noted between ICR and child's cognitive score at 20–24 months, whereas no positive association with ICR and growth was detected. Iodine sufficiency may be important for child's cognitive development in this setting.

Keywords
Child development
Education
Growth
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Iodine is necessary for the production of thyroid hormones thyroxine and triiodothyronine that are needed for normal human growth and development[1]. Data from 2011 indicated that about 41 million newborns per year worldwide were unprotected from iodine deficiency-induced consequences of brain damage[2]. Globally, insufficient intake of iodine is still one of the most common micronutrient deficiencies[3], despite it being one of the most preventable causes of impaired cognitive development in children[4]. In line with this, a recent cluster-randomised controlled trial from Ethiopia showed improved mental development among children aged 20–29 months after receiving iodised salt[5]. Notably, iodine deficiency among children <5 years is linked not only to poor mental development but also to stunted growth[1,6]. However, in their systematic review, Farebrother *et al.*[7] questioned whether prenatal iodine

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repletion would increase infant growth, highlighting the need for further research. Yet, discussions on the aetiology of childhood stunting involving nutrition seldom include iodine deficiency[8]. Contrary to many other nutrient deficiencies, iodine deficiency occurs in both developed and developing countries[8]. Several countries have implemented the use of iodised salt as a cheap public health measure to prevent iodine deficiency disorders[4]. A UNICEF report from 2015 indicated that about 75 % of households worldwide use iodised salt[9]; henceforth the number of iodine-deficient countries has decreased from 110 in 1993 to 15 in 2016[10]. Moreover, a recent report from the Iodine Global Network database indicated that 129 out of 197 countries have mandatory legislation for the iodisation of at least household/table salt or salt for food processing[11].

With the use of urine iodide concentration (UIC, a marker of iodine intake), according to the WHO, the threshold for iodine deficiency among children is UIC < 100 μg/l (<0·79 μmol/l)[12]. Using this threshold, Harika *et al.*[13] reported large variations in the prevalence of iodine deficiency among children aged 0–19 years in Ethiopia (86 %), Nigeria (59 %) and South Africa (15 %).

In Uganda, >95 % of the households consume iodised salt[14]. In 2002, a study on the severity of iodine deficiency disorders showed that about 30 % of the Ugandan general population had been diagnosed with goitre, indicating a severe public health problem[14]. However, the current prevalence of iodine deficiency is unclear since few surveys provide accurate estimates[10]. Specifically, Uganda's districts bordering the Democratic Republic of Congo, with high mountainous terrains, deep valleys, volcanic soils and abundant rainfall, are endemic for iodine deficiency[15]. To illustrate this, Ehrenkranz *et al.*[15] found that the prevalence of iodine deficiency was 21 and 23 % among newborns in the districts of Kabale and Kisoro, respectively.

Although several studies have investigated the adverse outcomes of iodine deficiency on child health[4,16,17], few studies have examined the association of iodine status with development and growth, especially in Sub-Saharan Africa[2,18]. Moreover, health promotion interventions for children have usually overlooked iodine's importance for child health outcomes[19].

We performed a randomised trial to test the effect of a 6-month intervention, consisting of nutrition, stimulation and hygiene education among mothers of children aged 6–8 months in Uganda, on growth and development[20]. Whereas this education intervention did not improve child growth at the age of 20–24 months, cognitive, language and motor development improved. Given the possible importance of *inter alia* an adequate iodine intake on both child development outcomes and growth, we here used data from our follow-up of this trial. This presented a unique opportunity to study development and growth over time, that is, when the children were 20–24 and 36 months, and specifically to (i) evaluate the effects of the intervention on iodine intake (measured as urine iodide excretion) and (ii) perform a secondary analysis to search for associations between urine iodide excretion and child development as well as growth.

Methods

Study area and participants

This is a secondary analysis of a follow-up study of our open, cluster-randomised education intervention regarding nutrition, stimulation and hygiene among impoverished mothers of children aged 6–8 months in the Kisoro and Kabale districts of south-western Uganda; details can be found elsewhere[20]. We report the data according to CONSORT guidelines.

Randomisation and allocation to study groups

The randomisation procedure of the original trial is detailed elsewhere[20]. In brief, ten clusters were first obtained (i.e., sub-counties) from the two study districts by simple random sampling before they were randomised to either intervention (*n* 5) or control (*n* 5). Second, all the villages in each cluster were listed alphabetically and assigned numbers. Using computer-generated random numbers, villages whose assigned number matched with the random numbers were chosen. We accounted for an intra-cluster correlation (a measure of relatedness of clustered data) for a linear growth of 0·01[21]. We finally enrolled 511 mother–child pairs in the original study, and they were randomised to an intervention (*n* 263) or a control (*n* 248) group. The intervention group received the nutrition, hygiene and stimulation education in addition to routine healthcare, while the control group received only routine healthcare.

The child had to be 20–24 months of age during January–May 2015 in order to be included in the current follow-up study, since developmental milestones at this age may predict IQ at 5–6 years when children are about to start school[22]. Out of the 511 children in the original trial, 501 were 20–24 months at the time of this follow-up study. Among these 501 children, 155 were randomly selected to participate in the current study. The primary outcome in this follow-up study was cognitive development assessed with the Bayley Scales of Infant and Toddler Development–III (BSID-III) at 36 months. To detect a clinically relevant difference between the two study groups in the BSID-III cognitive composite score at 36 months of 0·5 SD (corresponding to 7·5 points) with a power of 0·8 and α of 0·05, sixty-three children per group were required. To account for dropouts, a total of 155 children were included. Among these 155 children, we randomly selected seventy-seven from the intervention group and seventy-eight from the control group.

Data were collected when the children were 20–24 and 36 months. The data collection teams were masked to group allocation and never interacted with the team that delivered the education intervention in the original trial.

Delivery and content of education intervention in the original trial

The education intervention lasted 6 months starting when the children were between 6 and 8 months of age and has been described elsewhere[20]. Mothers in the intervention group were periodically followed and reminded of the intervention activities. Following the end of intervention period, eight booster education sessions were conducted after every 3 months until the children were 36 months. The intervention education teams emphasised nutrition, child stimulation and hygiene in these booster sessions.

Briefly, education intervention was delivered to groups of mothers by a team of nutrition educators (bachelor education in nutrition) following a nutrition education curriculum based on the ten guiding principles of complementary feeding[23]. The nutrition educators demonstrated breastfeeding practices and cookery. The mothers were advised: to start complementary feeding to their children with nutrient-rich foods while breastfeeding continued; to increase the number of feeds to 3–4 times a day; and to provide nutritious snacks between main meals. The mothers were also encouraged to practice responsive feeding and allow the children to feed themselves. The importance of oral hygiene and sanitation was given special emphasis.

Mothers together with the intervention team engaged in specific play activities and toys that could be useful in developing each of the development domains (cognitive, language and motor). The stimulation intervention was based on social-cognitive learning theory, emphasising the benefits of stimulation practices[24]. Mothers in the intervention group also met at monthly intervals to practice what they had learnt, thereby empowering them and ensuring compliance to the intervention[20].

Collection of development and growth data

We have detailed our data collection procedures for child development and anthropometric measurements[20]. In case of child illness, data collection was postponed. Three assistants with a bachelor degree in psychology performed child development assessments, while two with a bachelor degree in nutrition collected anthropometric data. Assessments were administered in the local language and conducted in hired rooms in the villages without distractions to minimise interruptions. To promote reliability, child development assessments were administered first, followed by anthropometric measurements, and then urine sampling. BSID-III and the Ages and Stages Questionnaire (ASQ) were used. BSID-III is known to be the most comprehensive child development measure for children up to 3·5 years and has been adapted in similar settings[25]. Unfamiliar items in the BSID-III stimulus and picture booklets were replaced with familiar objects in the Ugandan context; for example, apples were replaced with tomatoes, and a vacuum cleaner was replaced with a mop. Replacement items were chosen based on their size, colour and shape to maintain functional equivalence with the original stimuli. ASQ is a parent/caregiver-completed screening scale with excellent psychometric properties, which capture and establish a wide range of adaptive behaviours, and has been previously used in similar settings[26]. For mothers who could not read the translated ASQ tool in the local language, the assessments were conducted together with our blinded data collection team. This team would read ASQ questions to the mothers and then they would score the results together. Notably, few women could not read the local language; we registered only five (3 %) mothers out of 155. Both tools were used because ASQ assesses the social-emotional abilities of the child, which were not included in our BSID-III. In addition, ASQ is used to evaluate a range of adaptive behaviours not obtained with BSID-III and which the child may not readily perform during testing. Inter-observation agreement between the teams was good, indicated by an intra-class correlation coefficient of 0·75 ($P < 0·001$) for BSID-III and 0·79 ($P < 0·001$) for ASQ.

Nutritional status was evaluated using weight and length following standard procedures and calibrations recommended by WHO[27]. Weight (to the nearest 0·1 kg) was measured with a Seca scale model 881, whereas height (recumbent at age 20–24 and standing at age 36 months) was measured (to the nearest 0·1 cm) with a length board/stadiometer (Seca). The date of birth was obtained from the child's health card. These anthropometric data were converted to *z*-scores for height-for-age (HAZ), weight-for-age (WAZ), weight-for-height (WHZ), head circumference and mid-upper arm circumference using the WHO Anthro (version 3.2.2) software[28]. Undernutrition (stunting, underweight and wasting) was defined as a *z*-score <–2 SD from the median of the WHO reference standards for HAZ (stunting), WAZ (underweight) or WHZ (wasting), respectively[28].

Urine iodide and creatinine concentrations

We collected 155 and 148 samples of morning spot urine (volumes ranged from 2·5 to 4 ml) at 20–24 and 36 months, respectively. These samples were collected by a graduate student of laboratory technology using small containers and were transferred to tubes and kept at 4 °C for no more than 24 h before being frozen at –20°C. They were then shipped on dry ice to Oslo University Hospital for analysis at the Department of Medical Biochemistry. We measured the concentration of anion (oxidised form of iodine), which is iodide. Briefly, urine iodide was analysed by a colorimetric method based on ammonium persulfate digestion prior to the Sandell–Kolthoff reaction, as described by Ohashi *et al.*[29] and with an analytical CV of 6 % at 0·9 μmol/l.

Creatinine in urine was measured with enzymatic colorimetry using Cobas 6000 (Roche; CV 3 %). Urine was collected as spot urine samples, which were passed out when the children's bladders were full. It was not feasible to collect diurnal urine samples. UIC was corrected for

differences in water intake and, hence, urine dilution and concentration by calculating the individual urine iodide/creatinine ratio (ICR), which was used as a measure of iodine status in addition to UIC[30].

Measurements of iodine concentration in drinking water

To determine the concentration of iodine in drinking water, twenty randomly selected samples (ten from intervention villages and ten from control villages) were collected from the following sources of water: protected springs (*n* 4), unprotected springs (*n* 4), free-flowing springs (*n* 3), ponds (*n* 2), gravity (i.e., tap water, *n* 3) and swamp water (*n* 4). Iodine concentrations were analysed by Vestfold Lab Ltd. using the ISO 17294-2 method (level of detection 0·5 µg/l). Iodine was extracted by an aqueous solution of tetramethylammonium hydroxide (TMAH; 0·2–1·0 g sample in 1 ml TMAH and 5 ml H_2O). Extraction was carried out at a temperature of 90°C for 3 h. After cooling, the sample was diluted, the liquid phase was separated and prepared for measurement by ICP-MS (after the addition of an internal standard).

Statistical analyses

Values are reported as mean (95 % CI), or median with IQR, as appropriate. Differences between the study groups in concentrations of urine compounds were tested by Mann–Whitney *U* tests for each time-point since the data was not normally distributed. For the secondary analyses, we used mixed models to investigate the effect of urine iodide on growth and development outcomes in the intervention and control groups separately. An individual child was set as a random identifier; time and ICR/UIC as fixed variables. We adjusted for baseline values (obtained when the children were 6–8 months) of the outcomes of interest. To investigate whether the effect of urine iodide on different outcomes changed between 20–24 and 36 months, we included an interaction term 'time × UIC' and 'time × ICR'. The association of iodine with child development and growth was expressed as a regression coefficient with 95 % CI and its corresponding *P*-value. The analyses were performed with Stata/se 14 (StataCorp. 2015) and IBM SPSS Statistics, version 22.0 (IBM Corp.).

Results

Inclusion and characteristics of participants in the follow-up study

Figure 1 shows the inclusion criteria for both the original trial cohort and the follow-up study cohort. The characteristics of the original trial cohort and the follow-up study cohort are shown in Table 1. Anthropometry and BSID-III scores

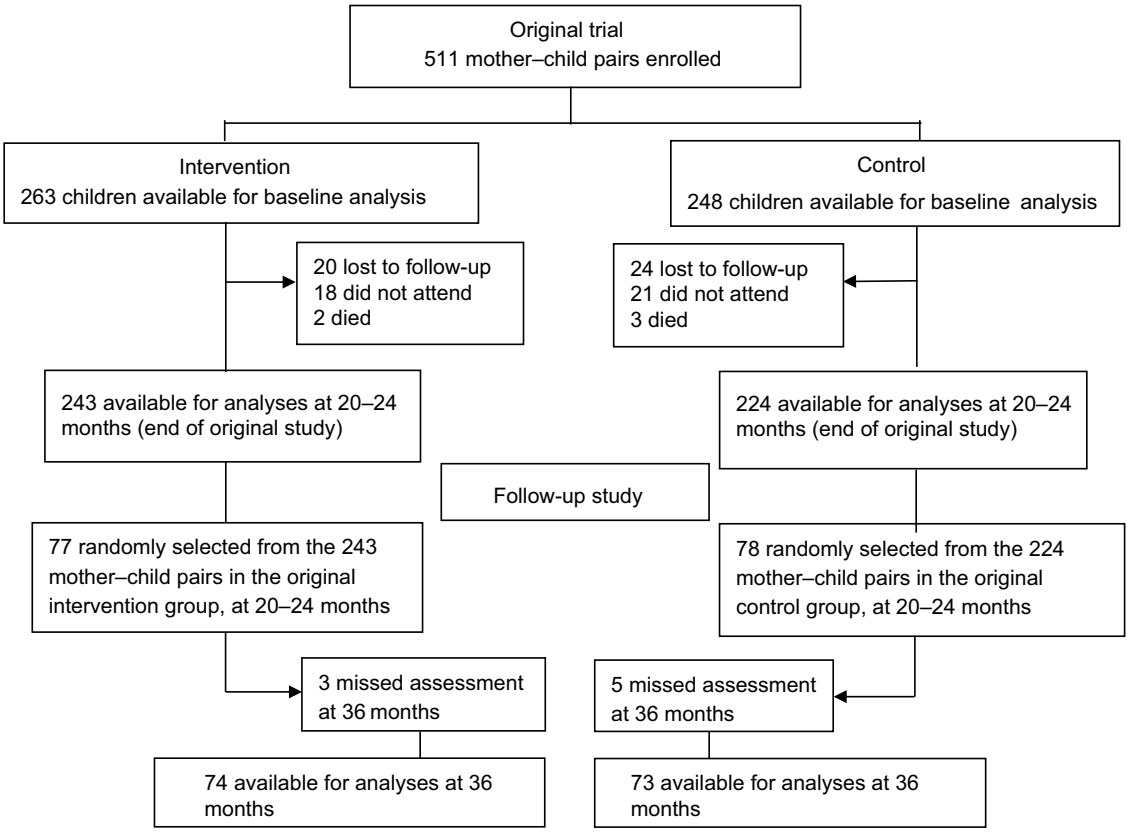

Fig. 1 Flowchart of inclusion process

Table 1 Population characteristics of the original trial cohort and the follow-up study cohort

	Original trial cohort (6–8 months)				Follow-up study cohort (20–24 months)			
	Intervention (*n* 263)		Control (*n* 248)		Intervention (*n* 77)		Control (*n* 78)	
Characteristics	*n*	%	*n*	%	*n*	%	*n*	%
Children								
Males	139	52·9	123	49·6	44	57·1	41	52·6
Females	124	47·1	125	50·4	33	42·9	37	47·4
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Child's age (months)	7·4	0·8	7·3	0·9	21·4	1·0	21·2	1·0
Maternal data								
Education (years)	4·9	2·8	4·9	2·8	5·5	2·5	5·0	2·6
Age (years)	26·1	5·8	26·8	6·3	26·2	6·1	27·4	6·4
Number of children per mother	3·4	2·2	3·3	2·2	3·4	2·2	3·3	2·2
Household data								
Household head's age (years)	31·3	7·7	33·4	10·7	30·2	7·3	33·1	10·9
Household head's education (years)	6·4	3·1	5·9	3·1	6·6	3·3	6·5	3·4
Household size (*n*)	5·5	2·1	5·5	2·1	5·7	2·2	5·8	2·2
Household poverty score*	47·8	11·7	47·6	11·4	49·0	11·6	46·3	12·3
Sanitation composite score	7·2	1·9	7·3	1·9	7·0	1·8	7·1	1·9

*Poverty score card[46].

Table 2 Anthropometry and Bayley Scales of Infant and Toddler Development–III (BSID-III) scores for the original trial cohort and the follow-up study cohort

	Original trial cohort				Follow-up study cohort			
	Intervention (*n* 240–263)*		Control (*n* 212–248)*		Intervention (*n* 73–77)*		Control (*n* 74–78)*	
	n	%	*n*	%	*n*	%	*n*	%
Child growth at 6–8 months								
Stunting†	55	20·9	70	28·0	14	18·9	28‡	38·4
Underweight†	25	9·5	36	14·5	7	9·5	8	11·0
Wasting†	12	4·6	12	4·8	3	3·9	2	2·6
Child growth at 20–24 months								
Stunting†	142	49·3	146	50·7	32	41·6	46	59·0
Underweight†	22	8·9	27	12·1	6	7·8	8	10·3
Wasting†	1	0·4	2	0·9	3	3·9	2	2·6
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
BSID-III scores at 6–8 months								
Cognitive	102·1	12·9	103·4	13·8	101·3	11·9	104·4	14·5
Language	103·5	14·4	100·2	14·1	102·4	16·2	101·8	15·2
Motor	104·9	13·8	104·4	14·7	108·8	14·6	106·6	15·8
BSID-III scores at 20–24 months								
Cognitive	114·9	21·3	99·3‡	17·1	117·8	20·9	101·6‡	19·1
Language	98·3	14·3	88·4‡	9·1	100·3	12·9	89·0‡	9·3
Motor	113·7	18·9	99·1‡	14·3	113·8	16·1	100·0‡	15·5

*Variation in *n* is due to missing data.
†Based on *z*-score values < 2 SD of the median of reference population.
‡Significant differences between the intervention and control groups.

obtained when the children were 20–24 months are shown in Table 2. The corresponding data obtained when the children were 36 months have recently been reported[32]. The intervention group in the follow-up study cohort had significantly fewer cases of stunting than the controls when they were 20–24 months. Moreover, the intervention groups in both the original trial cohort and the follow-up study cohort had significantly higher BSID-III scores at 20–24 months.

Urine iodide concentrations at 20–24 and 36 months

According to the criteria proposed by Andersson *et al.*[2], adequate UIC for small children ranges from 100 to 199 μg/l (0·79–1·60 μmol/l). Median UIC for both study groups at 20–24 and 36 months were within this range (Table 3). At 20–24 months, 20·8 and 21·8 % of children in the intervention and control groups, respectively, had a UIC <100 μg/l (0·79 μmol/l), whereas 35·1 and 52·6 %, respectively, had

Table 3 Urine iodide concentration (UIC) and iodide/creatinine ratio (ICR) of the follow-up study groups

		Intervention (*n* 75–77)*		Control (*n* 73–78)*		
Variable range	Child's age (months)	Median	IQR	Median	IQR	*P*-value**
UIC, μmol/l†	20–24	1·50	0·20–5·50	1·60	0·20–5·51	0·11
	36	1·20	0·20–5·50	1·60	0·20–5·52	0·08
ICR	20–24	4·9	1·0–9·6	2·7	1·0–10·1	0·03
	36	4·2	1·1–10	4·6	1·1–9·2	0·34
Creatinine (mg/dl)†	20–24	22·4	3·1–146·2	18·9	3·5–158·1	0·69
	36	27·0	2·0–121·5	24·4	3·0–112·1	0·35

*Variation in *n* is due to some children not completing all the tests.
†1 μmol/l of UIC corresponds to 127 μg/l, whereas 1 mg/dl of creatinine corresponds to 88·4 μmol/l.
***P*-value for the difference between study groups at each time-point.

Table 4 Associations between urine iodide/creatinine ratio (ICR) and child developmental scores of the two study groups

		Intervention (*n* 75–77)*				Control (*n* 73–78)*			
Bayley Scales of Infant and Toddler Development–III (BSID-III)	Child's age (months)	*R*†	95 % CI	*P*-value***	$P_{interaction}$#	*R*†	95 % CI	*P*-value***	$P_{interaction}$#
Cognitive	20–24	1·76	0·19, 3·33	0·028	0·106	0·51	−0·10, 2·03	0·505	0·765
	36	−0·32	−2·29, 1·65	0·753		0·17	−1·55, 1·88	0·849	
Language	20–24	0·31	−0·81, 1·43	0·584	0·573	−0·0	−0·95, 0·87	0·931	0·897
	36	−0·21	−1·61, 1·19	0·773		0·05	−0·98, 1·07	0·920	
Motor	20–24	0·62	−0·78, 2·01	0·384	0·081	0·32	−1·10, 1·84	0·678	0·369
	36	−1·39	−3·15, 0·37	0·121		1·39	−0·35, 3·12	0·117	
Ages and Stages Questionnaire (ASQ)									
Communication	20–24	−0·19	−1·19, 0·81	0·711	0·933	0·04	−1·10, 1·27	0·954	0·814
	36	−0·26	−1·52, 1·01	0·688		0·26	−1·10, 1·63	0·704	
Gross motor	20–24	0·36	−0·35, 1·07	0·316	0·647	−1·27	−2·34, 0·21	0·020	0·180
	36	0·09	−0·80, 0·99	0·835		−0·15	−1·32, 1·01	0·795	
Fine motor	20–24	0·41	−0·43, 1·26	0·340	0·858	0·15	−0·10, 1·29	0·801	0·837
	36	0·54	−0·55, 1·62	0·331		0·33	−0·95, 1·61	0·614	
Problem-solving	20–24	0·77	−0·11, 1·66	0·087	0·358	0·09	−1·01, 1·18	0·878	0·545
	36	0·10	−1·03, 1·22	0·867		0·60	−0·61, 1·82	0·333	
Personal/social	20–24	0·23	−0·61, 1·07	0·586	0·072	−0·30	−1·16, 0·55	0·489	0·927
	36	−1·01	−2·08, 0·05	0·063		−0·24	−1·20, 0·72	0·622	

*Variation in *n* is due to incomplete data.
†Values are regression coefficients (*R*) adjusted for baseline scores.
***Mixed-effects linear regression *P*-values of the association between ICR and BSID-III/ASQ scores.
#*P*-value for the difference between two time-points' regression coefficients.

a UIC >200 μg/l (1·60 μmol/l). The corresponding values at 36 months were 26·0 and 31·6 % with a UIC <100 μg/l (0·79 μmol/l), and 40·3 and 52·6 % with a UIC >200 μg/l (1·60 μmol/l) in the intervention and control groups, respectively. No child had a severe iodine deficiency defined as UIC < 20 μg/l (0·16 μmol/l) at either time-point.

ICR was nearly twice as high in the intervention compared with the control group at 20–24 months ($P = 0·03$), whereas no significant difference was found at 36 months (Table 3). We did not detect any significant differences in UIC, neither between the two study groups nor between the two time-points of assessment (Table 3).

Associations between iodine status and child's development outcomes

In both the original trial and the follow-up study, we found that the intervention led to better developmental outcomes. We here show that the intervention group had higher ICR at 20–24 months compared with the controls. Therefore, we next examined whether the ICR was associated with any of the development outcomes in the two study groups. Table 4 shows that ICR was positively associated with BSID-III cognitive scores at 20–24 months in the intervention group, but not with any other developmental outcome in either the intervention or control group when adjusting for baseline values. None of the ASQ scores were significantly associated with ICR either at 20–24 or 36 months in the two study groups except that ICR was negatively associated with ASQ gross motor score ($P = 0·020$) among the controls at 20–24 months (Table 4). The associations of ICR on BSID-III and ASQ development outcomes did not differ between the two time-points (i.e., $P > 0·05$ for the interaction term).

UIC was positively associated with BSID language development scores at 20–24 months in the control group, but negatively associated at 36 months (Table 5). Thus, we found a significant interaction effect between time and UIC

Table 5 Associations between urine iodide concentration (UIC) and child developmental scores of the two study groups

Bayley Scales of Infant and Toddler Development–III (BSID-III)	Child's age (months)	Intervention (*n* 75–77)*				Control (*n* 73–78)*			
		R†	95 % CI	*P*-value***	$P_{interaction}$#	*R*†	95 % CI	*P*-value***	$P_{interaction}$#
Cognitive	20–24	−2·68	7·52, 2·16	0·278	0·356	0·58	2·39, 3·57	0·70	0·266
	36	0·31	3·73, 4·36	0·880		−1·74	−4·55, 1·08	0·23	
Language	20–24	1·63	−1·74, 5·01	0·342	0·434	1·77	0·07, 3·46	0·041	0·001
	36	−0·13	−2·96, 2·70	0·928		−2·05	−3·63, −0·46	0·011	
Motor	20–24	0·92	−3·36, 5·19	0·42	0·821	1·59	−1·39, 4·57	0·296	0·037
	36	1·57	−2·03, 5·16	0·85		−2·75	−5·55, 0·05	0·054	
Ages and Stages Questionnaire (ASQ)									
Communication	20–24	−0·08	−3·11, 2·94	0·957	0·548	−0·73	−3·11, 1·66	0·551	0·632
	36	1·34	−1·41, 3·68	0·381		−1·52	−3·75, 0·71	0·183	
Gross motor	20–24	1·41	−0·69, 3·52	0·189	0·879	1·10	−1·04, 3·25	0·312	0·162
	36	−0·63	−2·60, 1·33	0·527		1·32	−0·48, 3·12	0·150	
Fine motor	20–24	0·01	−2·57, 2·59	0·996	0·166	0·16	−2·03, 2·35	0·885	0·087
	36	2·39	0·25, 4·52	0·029		−2·44	−4·49, −0·40	0·019	
Problem-solving	20–24	0·54	−2·18, 3·26	0·698	0·557	0·30	−1·83, 2·43	0·784	0·203
	36	1·62	−0·63, 3·89	0·159		−1·60	−3·59, 0·40	0·117	
Personal/social	20–24	0·27	−2·32, 2·86	0·837	0·793	−0·69	−2·35, 0·97	0·413	0·518
	36	−0·18	−2·36, 1·99	0·869		−1·45	−3·03, 0·14	0·074	

*Variation in *n* is due to incomplete data.
†Values are regression coefficients (*R*) adjusted for baseline scores.
***Mixed-effects linear regression *P*-values of the association between UIC and BSID-III/ASQ scores.
#*P*-value for the difference between two time-points' regression coefficients.

Table 6 Associations between urine iodide/creatinine ratio (ICR) and child growth *z*-scores of the two study groups

Growth	Child's age (months)	Intervention (*n* 75–77)*				Control (*n* 73–78)*			
		R†	95 % CI	*P*-value***	$P_{interaction}$#	*R*†	95 % CI	*P*-value***	$P_{interaction}$#
Height-for-age *z*-scores	20–24	0·03	−0·04, 0·11	0·381	0·196	−0·05	−0·12, 0·02	0·167	0·214
	36	−0·05	−0·14, 0·04	0·338		0·02	−0·06, 0·10	0·625	
Weight-for-age *z*-scores	20–24	−0·02	−0·07, 0·03	0·371	0·735	−0·01	−0·06, 0·05	0·829	0·262
	36	−0·01	−0·07, 0·05	0·776		0·04	−0·02, 0·10	0·174	
Weight-for-height *z*-scores	20–24	−0·03	−0·09, 0·03	0·324	0·415	0·01	−0·05, 0·08	0·688	0·939
	36	0·01	−0·06, 0·08	0·796		0·01	−0·06, 0·08	0·780	
Head circumference *z*-scores	20–24	0·06	−0·00, 0·11	0·051	0·027	−0·02	−0·09, 0·04	0·503	0·352
	36	−0·05	−0·12, 0·03	0·197		−0·07	−0·14, 0·00	0·055	
Mid-upper-arm *z*-score	20–24	0·01	−0·05, 0·06	0·846	0·772	0·00	−0·06, 0·06	0·905	0·802
	36	−0·01	−0·07, 0·06	0·829		0·02	−0·05, 0·08	0·651	

*Variation in *n* is due to incomplete data.
†Values are regression coefficients (*R*) adjusted for baseline scores.
***Mixed-effects linear regression *P*-values of the association between ICR and growth scores.
#*P*-value for the difference between two time points' regression coefficients.

on BSID language development in the control group. Furthermore, UIC was positively associated with ASQ fine motor scores at 36 months in the intervention group, but negatively associated among the controls.

Associations between iodine status and growth outcomes

None of the anthropometrical markers of growth was associated ($P > 0·05$) with ICR at the two time-points (Table 6). However, a significant interaction effect of ICR and time for head circumference *z*-score was found in the intervention group. Moreover, we found no significant associations between UIC and growth outcomes at 20–24 and/or at 36 months, except that at 20–24 months the *z*-scores for head circumference ($P = 0·006$) and weight-for-height ($P = 0·047$) were negatively associated with UIC in the control group (Table 7). Furthermore, a significant interaction effect was evident for UIC and time on *z*-scores for head circumference ($P = 0·033$).

Iodine concentration in drinking water

Since drinking water can be an important iodine source[32,33], we measured iodine concentrations in sources of drinking water from randomly selected ten intervention and ten control villages. The median (IQR) iodine concentrations were 17·5 (3·0–28·8) and 11·0 (2·5–30·1) μg/l for the intervention and control sources ($P = 0·78$), respectively. Hence, children

Table 7 Associations between urine iodide concentration (UIC) and child growth *z*-scores of the two study groups

Growth	Child's age (months)	Intervention (*n* 75–77)*				Control (*n* 73–78)*			
		R†	95 % CI	*P*-value***	$P_{interaction}$#	*R*†	95 % CI	*P*-value***	$P_{interaction}$#
Height-for age	20–24	–0·02	–0·25, 0·21	0·858	0·900	0·02	–0·12, 0·16	0·778	0·450
z-scores	36	–0·00	–0·19, 0·89	0·987		–0·05	–0·18, 0·08	0·423	
Weight-for-age	20–24	–0·06	–0·21, 0·09	0·424	0·568	–0·07	–0·17, 0·04	0·208	0·816
z-scores	36	–0·00	–0·12, 0·12	0·967		–0·05	–0·15, 0·04	0·300	
Weight-for-height	20–24	–0·07	–0·25, 0·10	0·419	0·266	–0·12	–0·24, –0·00	0·047	0·251
z-scores	36	0·06	–0·08, 0·20	0·399		–0·03	–0·14, 0·09	0·652	
Head circumference	20–24	0·06	–0·13, 0·24	0·59	0·734	–0·18	–0·30, –0·05	0·006	0·033
z-scores	36	0·01	–0·14, 0·16	0·18		0·01	–0·11, 0·13	0·876	
Mid-upper-arm	20–24	0·09	–0·07, 0·26	0·256	0·232	–0·07	–0·18, 0·05	0·273	0·924
z-scores	36	–0·04	–0·17, 0·10	0·588		–0·06	–0·17, 0·05	0·300	

*Variation in *n* is due to incomplete data.
†Values are regression coefficients (*R*) adjusted for baseline scores.
***Mixed-effects linear regression *P*-values of the association between UIC and growth scores.
#*P*-value for the difference between two time-points' regression coefficients.

in both study groups apparently received similar iodine content from drinking water.

Discussion

To our knowledge, the current study is apparently the first to focus on iodine status and its associations with child development and growth outcomes longitudinally in a rural setting and conducted in a low-income country.

The majority of children in the current cohort had sufficient iodine intake (i.e., UIC > 100 µg/l) both at 20–24 and 36 months of age even without specific iodine supplements. We also found that the median UIC in both study groups were within the recommended range as proposed by Andersson *et al.*[2]. This could be attributed to the Ugandan policy on the levels of iodine fortification of salt (95 % Ugandan households consuming iodised table salt)[14]. However, we found that about one-fifth of children in both study groups had low UIC at 20–24 months, and this fraction increased to about one-fourth and one-third in the intervention and control groups, respectively, after 1 year (at 36 months). Moreover, we found that the median ICR among intervention children was higher compared with the control group at 20–24 months, possibly indicating higher iodine consumption among children in the intervention group. In contrast, we did not detect any significant difference in median ICR at 36 months. Collectively, our data indicate that the intervention may have led to higher iodine consumption when the children reached the age of 20–24 months, but over time, the consumption possibly declined.

Interestingly, in our separate analyses of data from the two study groups, we found a positive association between ICR and BSID-III cognitive score in the intervention group at 20–24 months, and a negative association between ICR and ASQ gross motor score among the controls aged 20–24 months. Bougma *et al.* concluded in their systematic review that iodine deficiency has a substantial impact on mental development[1]. However, similar to our results, Bell *et al.*[34] in their systematic review also identified inconsistencies in the relationship between iodine and child development using global assessments. The aetiology behind a negative association between ICR and gross motor development at 20–24 months in the control group is unknown and may be due to chance. We do not know why the association for BSID-III language and motor scores was not positively associated with ICR as opposed to the cognitive scores in the intervention group at 20–24 months. Possibly, cognition has different development pathways compared with language and motor skills at an early age[35]. The lack of significant associations between iodine markers and outcomes at 36 months is consistent with that from other studies involving children, indicating that the main effects of iodine on development may be restricted to children aged ≤3 years[4,36,37]. Whereas Robinson *et al.* found no associations between ICR and executive function outcomes[37], the Ethiopian study focusing on the use of iodised salt reported similar findings to ours on the association of urine iodide and BSID-III cognitive outcomes[5].

We detected no significant association between ICR and the anthropometrical markers of child growth (HAZ, WAZ, WHZ, head circumference and mid-upper arm circumference) at 20–24-month and 36-month assessments. Our findings disagree with those obtained among Albanian and Moroccan children whose HAZ and WAZ increased after iodine supplementation[17]. Possibly this difference could be related to the fact that children in the current study were not supplemented with iodine. Notably, in the current study, children were using iodised table salt as part of Uganda's iodisation programme. However, our findings are comparable with studies from South Africa and Mexico that investigated the effects of increased iodine intake, which reported no effect on child growth outcomes[38,39]. These latter studies included micronutrient

supplements in their interventions. In contrast, we only provided an education intervention to mothers; no foods or supplements were provided at any time. Supporting our findings, a recent systematic review reported no clear evidence on the association between UIC and physical development; their findings rather identified likely increases in urinary iodine concentration[40].

We did not detect any significant difference in iodine content in the drinking water obtained from both intervention and control villages. Our results of the median range of water iodine concentrations are in line with those reported from, for example, Denmark[41], Austria, Spain[42], USA[43], and Australia and New Zealand[44]. Notably, WHO's recommended values for iodine in drinking water are not yet defined[45].

Several years of follow-up and including iodine measurements in both urine and drinking water samples constitute major strengths of the current study. We also adjusted for any effects of dilution or concentration of spot morning urine samples by reporting urine ICR. Moreover, since urine creatinine concentrations were similar between the study groups, there is no reason to believe that nitrogen intake (and thus protein intake) was different between the intervention and control groups. A major limitation of the current study was the lack of data on actual intake of iodine among children, neither from foods nor from drinking water as well as household table salt, as this was not included in the design of the original trial. For the same reason, we also lack data on baseline urine iodine concentrations when the children were recruited to the original trial. Also, this was a follow-up study of an original trial where the intervention did not target iodine intake as the primary outcome. Furthermore, the size of follow-up sample was smaller than in the original trial. Information from the mothers, such as ASQ being a maternal report, could possibly be biased. Since many comparisons were performed, we cannot rule out that some statistical associations may have occurred by chance alone.

In conclusion, the intervention only led to a positive association between ICR and child development outcomes at 20–24 months (measured as BSID-III cognitive scores). ICR was not associated with any growth outcomes neither at 20–24 months nor at 36 months. Our data suggest that iodine is important for child's mental development, at least for cognitive skills. Still, there is a need for further studies to establish the associations between iodine intake, child development and growth outcomes, especially in low-resource areas.

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