Nutrición Hospitalaria

Nutr Hosp. 2008;23(3):212-219
ISSN 0212-1611 • CODEN NUHOEQ
S.V.R. 318

Original

Zinc supplementation in infants with asymmetric intra uterine growth retardation; effect on growth, nutritional status and leptin secretion

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Abstract

Objectives: **To analyse the effect of zinc supplementation in growth and nutritional status of a homogeneous group of newborns with intra uterine growth retardation and asymmetric growth. The effect of changes of zinc status on growth and leptin serum concentrations was also analysed.**

Population and methods: **A double blind, randomised clinical trial was designed in order to detect differences in growth between zinc and placebo groups during the first 6 months of life. 31 infants were included either to the zinc group (n = 14) (38.8 ± 1.4 weeks GA, 2,171 ± 253 g body weight) or the placebo group (n = 17) (38.9 ± 1.1 weeks GA, 2,249 ± 220 g body weight). The zinc group received a supplement of 3 mg elemental zinc per day (as zinc sulphate).**

Results: **There were not significant differences between groups for anthropometric measurements through the study period. We found a significant effect of the study group, in hair zinc concentrations, but not in serum zinc concentrations; *post-hoc* comparisons for hair zinc revealed that there were significant differences between groups at 1, 2, and 6 months of age. Changes in serum and hair zinc concentrations from baseline to 6 months, showed significant correlations with changes in weight/age and length/age z-scores, in the supplement group. Changes in leptin serum concentrations during follow-up, showed significant correlations with changes in sum of 4 skinfolds and weight/age z-score, in the placebo group. Changes in hair zinc concentration through the study period showed significant correlations with changes in leptin serum concentrations from baseline to 6 months of follow-up.**

Conclusions: **In a homogeneous group of intra uterine growth retardation infants with asymmetric growth, 3**

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Recibido: 14-VI-2007.
Aceptado: 5-XI-2007.

SUPLEMENTACIÓN CON ZINC EN NIÑOS CON RETRASO DE CRECIMIENTO INTRA-UTERINO ASIMÉTRICO; EFECTO EN EL CRECIMIENTO, ESTADO NUTRICIONAL Y SECRECIÓN DE LEPTINA

Resumen

Objetivos: **Valorar el efecto de la suplementación con cinc en el crecimiento y estado nutricional de un grupo homogéneo de recién nacidos con retraso de crecimiento intra-uterino asimétrico. También se analizó el efecto de los cambios en el status del cinc en el crecimiento y las concentraciones séricas de leptina.**

Población y método: **Se diseñó un ensayo clínico randomizado y doble ciego, con el fin de detectar diferencias en el crecimiento entre los grupos recibiendo cinc o placebo, durante los seis primeros meses de vida. 31 niños fueron incluidos en el grupo cinc (n = 14) (38,8 ± 1,4 semanas edad gestacional, 2.171 ± 253 g peso) o grupo placebo (n = 17) (38,9 ± 1.1 semanas edad gestacional, 2.249 ± 220 g peso). El grupo cinc recibió un suplemento de 3 mg de cinc elemental por día (en forma de sulfato de cinc).**

Resultados: **No hubo diferencias significativas entre ambos grupos en cuanto a las medidas antropométricas a lo largo del período de estudio. Se observó un efecto significativo del grupo de estudio, en las concentraciones de cinc en el pelo, pero no en las concentraciones séricas de cinc; las comparaciones *post-hoc* para el cinc del pelo pusieron de manifiesto que había diferencias significativas entre los grupos, en los meses 1, 2 y 6 de edad. Los cambios en las concentraciones de cinc en el suero y en el pelo, desde el inicio del estudio hasta los 6 meses, mostraron correlaciones estadísticamente significativas con los cambios en peso/edad y longitud/edad (puntuación típica), en el grupo que recibió el suplemento de cinc. Los cambios en las concentraciones séricas de leptina durante el seguimiento, mostraron correlaciones estadísticamente significativas para la suma de 4 pliegues y para peso/edad (puntuación típica), en el grupo placebo. Los cambios en las concentraciones de cinc en el pelo mostraron correlaciones estadísticamente significativas con los cambios en las concentraciones séricas de leptina, desde el inicio del estudio hasta los 6 meses de seguimiento.**

Conclusiones: **En un grupo homogéneo de niños con retraso de crecimiento intra-uterino asimétrico, el suple-**

mg/day zinc supplementation do not show significant improvements in weight and length growth. Changes in zinc status were related with changes in weight and length during the first 6 months of life. Changes in leptin serum concentrations were related with changes in the anthropometric indices of body fat accretion.

(*Nutr Hosp.* 2008;23:212-219)

Palabras clave: *Zinc. Intrauterine growth retardation. Growth. Nutritional status. Leptin. Randomised trial.*

mento de cinc a una dosis de 3 mg/día, no origina mejora significativa en el crecimiento en peso y longitud. Los cambios en el status de cinc se relacionaron con los cambios en peso y longitud durante los 6 primeros meses de vida. Los cambios en las concentraciones séricas de leptina se relacionaron con los cambios en los índices antropométricos de acúmulo de grasa corporal.

(*Nutr Hosp.* 2008;23:212-219)

Key words: *Cinc. Retraso de crecimiento intrauterino. Crecimiento. Estado nutricional. Leptina. Estudio aleatorizado.*

Introduction

Gestational age and nutritional status at birth are important determinants of growth patterns in infancy. Infants who are born too small differ in many important ways from those born too soon. Intra uterine growth retardation (IUGR) represents 23.8% of all newborns in developing countries.[1] In the US, the frequency of IUGR in term newborns ranged from 2.3% at 38 gestational weeks to 1.1% at 40 gestational weeks.[2] Full term newborns with IUGR have small livers, and are expected to have a small reserve capacity for micronutrients stored in the liver. Zinc is among these micronutrients.

Low maternal zinc concentration during pregnancy may be associated with an increased risk of low birth weight.[3] A reduced level of zinc in low birth weight infants, might account for the growth failure in such children. Unfortunately, it is difficult to assess zinc status reliably, and any detrimental effect of zinc deficiency can only be established by zinc supplementation trials, assessed by growth indices and morbidity reduction as outcome variables.[4]

Evidence of the importance of zinc deficiency in child health has come from recent randomised, controlled trials of zinc supplementation.[5] Evidence for an effect of zinc supplementation on the growth of children came from a recent meta-analysis that concluded that zinc supplementation during childhood is responsible for a small but statistically significant effect on growth, particularly among growth-retarded children.[6] Most of the intervention trials studying the effect of zinc on growth or morbidity were performed in children older that 6 months of age, when the period of highest growth velocity has already passed. It was therefore hypothesized that earlier interventions might be more effective in preventing growth faltering among children at risk.[7]

Zinc supplementation in small for gestational age infants result in substantial reduction in infectious disease mortality.[8] In a community-based study conducted in India, it has also been observed that zinc supplementation has a beneficial impact on the incidence of diarrhoea and also weight gain among low birth weight infants.[9] Beneficial effects on growth were observed after zinc supplementation among low-birth-weight and small-for-gestational-age infants in Brazil[10] and Chile.[11]

Zinc plays an important role in appetite regulation. In rodents and humans, zinc deficiency decreases appetite, while zinc supplementation increases appetite.[12] The most widely postulated mechanism for zinc-induced changes in appetite is alteration in hypothalamic neurotransmitter metabolism.[13] Zinc status could influence the regulation of appetite and metabolism by influencing the leptin system. In adults, it has been observed that zinc restriction decreased leptin levels while zinc supplementation of zinc-depleted subjects increased circulating leptin levels.[14] Very early in life, it has been also observed that the normal 3 to 6% weight reduction that occurs during the first 4 postnatal days was associated with a 26% decrease in the plasma leptin level in healthy breastfed infants.[15]

The aim of our study was to analyse the effect of zinc supplementation in growth and nutritional status of a homogeneous group of newborns with IUGR and asymmetric growth retardation defined in terms of ponderal index. The effect of changes of zinc status on growth and leptin serum concentrations was also analysed.

Materials and methods

A longitudinal double blind, randomised clinical trial was designed in order to detect differences in growth between zinc and placebo groups during the first 6 months of life. The study was performed at the neonatal unit of the University Hospital of Zaragoza (Spain). Singleton newborns were eligible for inclusion in the study if they met entry criteria: Gestational age 38-41 weeks (Dubowitz method applied by a trained paediatrician), birth weight lower than 10th percentile for gestational age,[16] and ponderal index lower than 2.4 (weight in g x 100/length in cm^3). Infants with congenital malformations, asphyxia, or congenital infections, and those whose mothers had preeclampsia or intrahepatic cholestasis during pregnancy were excluded. Sample sizes were calculated for an assumed 5% difference in linear growth, a variance of 56.25 cm, a power of 80%, and a 1-side level of significance of 0.05. A total of 38 newborns were identified and enrolled in the study. Before enrollement, written, informed consent was obtained from each newborn's parents. The study was approved by the Ethical Committee of

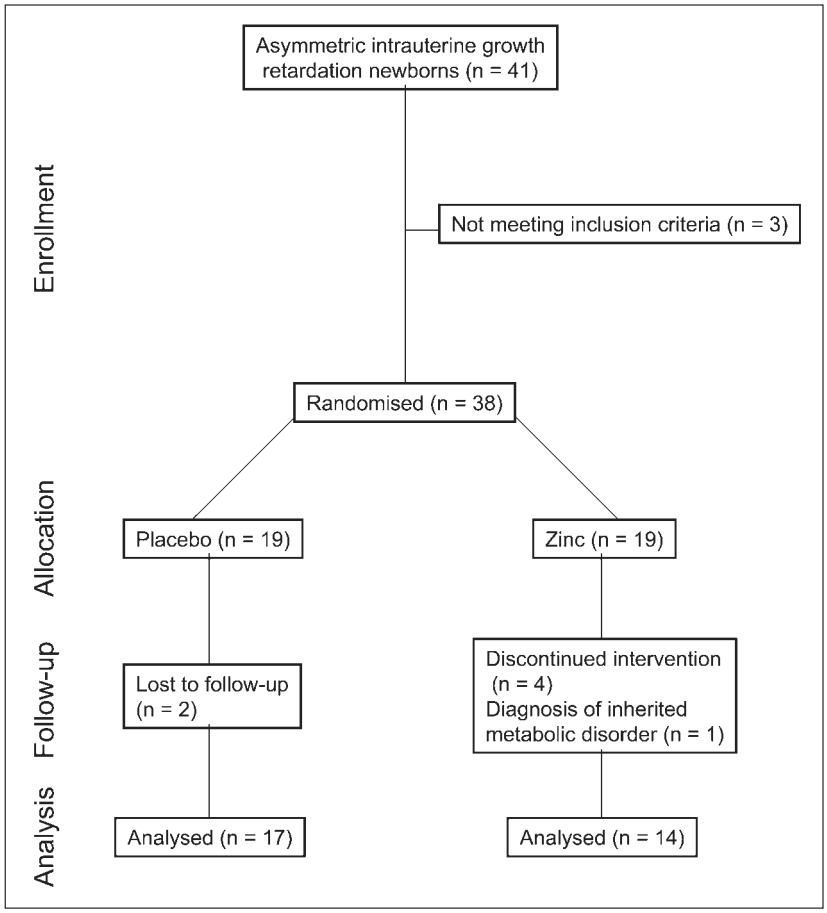

Fig. 1.—Flow chart of participants in each stage of the randomised trial.

the University Hospital "Lozano Blesa", Zaragoza (Spain). Parents knew they could withdraw their infants from the study at any time. During the follow-up, seven infants were excluded: 6 for lack of follow-up and one because of a diagnostic of an inherited metabolic disorder was done after 6 months of intervention (fig. 1). Of the remaining 31 infants 14 had been included in the zinc group (38.8 ± 1.4 weeks GA, 2,171 ± 253 g body weight) and 17 in the placebo group (38.9 ± 1.1 weeks GA, 2,249 ± 220 g body weight).

Newborns were randomly allocated to receive 3 mL/day of a solution with or without 3 mg elemental zinc (as zinc sulphate). Both solutions contained sucrose, flavours, and preservatives; were indistinguishable in both appearance and taste; and were prepared and coded by the Hospital Pharmacy. Investigators delivered a bottle containing a supply of 100 mL of the supplement to the houses of the participants and instructed the mother to give a daily dose to their infants, when possible between feedings, using a marked dropper or feeding spoon. Supplement (zinc or placebo) was started after the first blood sample. The codes of the supplements were unknown to both participants and study staff and were broken only after data editing and cleaning were completed. Randomisation was done during the 3 first days of life. Follow-up visits were performed at 1, 2, 4 and 6 months. Blood samples were also obtained in each visit. Information was collected on the household's socio-economic status, demographic characteristics and characteristics of the delivery (duration, complications). Information on infant feeding practices was collected every two weeks, and the infants were classified as either exclusively, partially breast-fed or not breast-fed at all. The age of introduction of complementary foods was also determined. No attempts were made to influence the mothers in their choice of infant feeding. Anthropometric measurements were performed in each follow-up visit. During the follow-up period, the infants were provided with the standard immunizations. Infants who required medical treatment were examined by study paediatricians following standard treatment protocols and were referred to appropriate health care facilities if necessary.

During each follow-up visit infant weight was measured to the nearest 10 g on a beam-balance scale (Seca

725, Seca, Hamburg, Germany) that was daily calibrated against standard weights. Recumbent lengths were measured to the nearest 0.1 cm on a length board. Head and arm circumferences were measured to the nearest millimetre with numeral insertion tapes. The same trained fieldworkers collected all data. The mean of two measurements was recorded as the observed value for all indices. Weight-for-age, length-for-age and weight-for-length standard deviation scores (SDS) were calculated with the use of ANTHRO software (CDC/WHO International Growth Reference). Ponderal index was also calculated: weight (g)/length (cm)^3.

Skinfold thicknesses were measured at the left side of the body to the nearest 0.1 mm with a Holtain skinfold caliper, at the following sites: 1) triceps, halfway between the acromion process and the olecranon process; 2) biceps, at the same level as the triceps skinfold, directly above the centre of the cubital fossa; 3) subscapular, about 20 mm below the tip of the scapula, at an angle of 45° to the lateral side of the body, and 4) suprailiac, about 20 mm above the iliac crest and 20 mm towards the medial line.[17] Intra-observer reliabilities were: 95.46% for biceps, 98.73% for triceps, 96.78% for subscapular, and 95.93% for suprailiac skinfold.

In each follow-up visit, non-fasting blood specimens were obtained by vein puncture, at 09:00 hours. For serum zinc determination, trace mineral free plastic syringes, stainless steel needles, and plastic tubes (Becton Dickinson Vacutainer Systems, Meylan, France) were used. Serum was separated a maximum of two hours after collection and after separation stored at -30 ºC until analysis. Zinc concentration was measured with an atomic absorption spectrophotometer (Video 11E, Thermo Jarrell Ash). Commercial serum with known concentrations of zinc was used as a measure of quality control.

Haemoglobin was determined in a haematological counter STKS model (Coulter Electronics, Inc., Hialeah, USA). Albumin, prealbumin, retinal binding protein, and trasnferrin serum concentrations were measured by kinetic immuno nephelometry (Array 360, Beckman).[18] The circulating serum leptin concentrations were determined with a commercially available human leptin RIA kit (Mediagnost, Tübingen, Germany). Serum concentrations of insulin like growth factor I (IGF-I) and insulin like growth factor binding protein 3 (IGFBP-3) were measured by radio immuno analysis (Nichols Institute Diagnostics, San Juan Capistrano, USA). Coefficients of variation for the

Table I
Changes in anthropometric measurements and indices through the study period

	Baseline	*1 month*	*2 months*	*4 months*	*6 months*
N					
Supplement	14	14	14	13	13
Placebo	17	17	17	17	17
Weight (g)					
Supplement	2,171 ± 253	3,306 ± 445	4,258 ± 577	5,850 ± 699	7,016 ± 718
Placebo	2,249 ± 220	3,323 ± 341	4,264 ± 395	5,765 ± 556	6,792 ± 590
Length (cm)					
Supplement	46.7 ± 1.7	50.5 ± 2.3	54.3 ± 2.1	60.5 ± 1.9	65.1 ± 2.1
Placebo	46.7 ± 1.9	50.7 ± 1.8	55.0 ± 1.9	60.7 ± 2.2	65.0 ± 2.6
Head circumference (cm)					
Supplement	32.2 ± 1.2	36.3 ± 1.4	38.0 ± 1.4	40.9 ± 1.3	42.9 ± 1.3
Placebo	32.4 ± 0.8	36.3 ± 1.0	38.2 ± 0.7	40.7 ± 0.7	42.9 ± 1.0
Arm circumference (cm)					
Supplement	9.2 ± 0.8	10.9 ± 1.1	12.0 ± 1.0	13.6 ± 1.0	14.4 ± 1.1
Placebo	9.0 ± 1.0	11.0 ± 0.9	12.1 ± 0.8	13.9 ± 0.8	14.5 ± 1.1
Sum 4 skinfolds (mm)					
Supplement	10.1 ± 1.8	18.2 ± 3.1	21.3 ± 2.1	24.7 ± 2.8	24.7 ± 4.3
Placebo	9.7 ± 2.3	17.6 ± 2.7	21.0 ± 4.0	24.1 ± 3.7	24.2 ± 4.1
Weight/age z-score					
Supplement	-2.5 ± 0.5	-1.4 ± 0.6	-1.1 ± 0.6	-0.9 ± 0.8	-0.9 ± 0.9
Placebo	-2.2 ± 0.4	-1.3 ± 0.5	-1.0 ± 0.4	-0.7 ± 0.4	-0.8 ± 0.4
Length/age z-score					
Supplement	-1.7 ± 0.7	-1.6 ± 0.8	-1.5 ± 0.8	-1.3 ± 0.7	-1.1 ± 0.8
Placebo	-1.6 ± 0.9	-1.4 ± 0.7	-1.1 ± 0.7	-0.8 ± 0.7	-0.7 ± 0.8
Weight/length z-score					
Supplement	-2.4 ± 0.2	-0.3 ± 0.6	0.0 ± 0.7	0.2 ± 0.8	-0.1 ± 0.7
Placebo	-2.3 ± 0.2	-0.4 ± 0.3	-0.2 ± 0.5	-0.2 ± 0.7	-0.4 ± 0.8

Not significant differences between groups through the study period.

Table II
Changes in serum and hair zinc concentrations through the study period

	Baseline	*1 month*	*2 months*	*4 months*	*6 months*
N					
Supplement	13	14	14	13	11
Placebo	15	15	15	14	16
Serum zinc (μg/dL)					
Supplement	134.5 ± 36.5	93.2 ± 26.5	116.4 ± 46.2	114.0 ± 44.7	107.3 ± 32.9
Placebo	130.4 ± 53.7	97.0 ± 50.3	108.2 ± 52.5	117.9 ± 51.5	105.6 ± 35.0
Hair zinc (μg/g)					
Supplement	219 ± 36	187 ± 49*	198 ± 67*	174 ± 50	197 ± 76*
Placebo	184 ± 60	148 ± 44	150 ± 47	152 ± 78	136 ± 48

* *Post-hoc* differences between supplement and placebo groups: $P < 0.05$.

RIA kits were the following: Leptin, intra-assay < 5%, and inter-assay 7.6%; IGF-I, intra-assay 2.4 to 3%, inter-assay 5.2 to 8.4%; and IGFBP-3, intra-assay 3.4 to 8% and inter-assay 5.3 to 6.3%.

To measure hair zinc concentrations, the hair samples were obtained near at the occipital region with stainless steel scissors. The samples were washed with neutral shampoo and then with deionised water; samples were dried at 60º for 12 h. Thereafter, they were digested with ultra pure nitric acid and hydrogen peroxide and diluted in distilled deionised water. Zinc concentration was measured by inductively coupled plasma mass spectrometry, by using am Emision Perkin Elmer Plasma 40 at a wave length of 214 nm. Mean coefficient of variation was 1.6% and the limit of experimental detection 0.0018 mg/mL.

Statistical analysis was performed by using the Statistical Package for Social Science version 11.5

Table III
Changes in biochemical data through the study period

	Baseline	*1 month*	*2 months*	*4 months*	*6 months*
N					
Supplement	14	13	13	13	11
Placebo	17	15	16	16	16
Haemoglobin (g/dL)					
Supplement	19.5 ± 2.1	11.9 ± 1.5	11.4 ± 0.9	12.6 ± 1.4	12.2 ± 1.3
Placebo	19.5 ± 1.9	12.1 ± 2.0	11.8 ± 2.0	11.9 ± 1.0	12.3 ± 1.1
Albumin (mg/dL)					
Supplement	3,204 ± 664	3,341 ± 466	3,649 ± 505	3,829 ± 493	3,887 ± 288
Placebo	3,310 ± 418	3,378 ± 499	3,883 ± 533	3,911 ± 589	4,079 ± 667
Prealbumin (mg/dL)					
Supplement	7.9 ± 1.5	11.2 ± 1.6	14.0 ± 1.8	15.9 ± 3.6	17.5 ± 4.4
Placebo	8.4 ± 3.0	11.2 ± 2.3	14.8 ± 3.1	17.3 ± 3.7	20.1 ± 2.6
Retinol binding protein (mg/dL)					
Supplement	0.95 ± 0.99	2.65 ± 1.1	4.3 ± 4.6	2.7 ± 2.3	2.6 ± 1.15
Placebo	1.00 ± 1.02	2.10 ± 1.12	2.90 ± 2.12	2.63 ± 4.02	2.80 ± 2.66
Transferrin (mg/dL)					
Supplement	189.5 ± 37.6	179.9 ± 25.2	240.5 ± 34.0	263.9 ± 37.7	271.4 ± 50.8
Placebo	201.1 ± 71.3	194.3 ± 61.0	250.0 ± 44.8	273.4 ± 53.0	268.1 ± 55.7
Leptin (ng/mL)					
Supplement	0.46 ± 0.54	-	-	3.62 ± 2.63	3.45 ± 1.34
Placebo	0.53 ± 0.55	-	-	2.79 ± 1.54	2.70 ± 2.27
IGF-I (nmol/L)					
Supplement	4.1 ± 1.5	11.3 ± 2.6	12.0 ± 2.9	12.4 ± 5.4	10.9 ± 3.9
Placebo	5.1 ± 0.8	10.7 ± 2.8	11.6 ± 2.8	11.5 ± 3.6	12.2 ± 3.4
IGFBP-3 (μg/mL)					
Supplement	0.56 ± 0.37	1.40 ± 0.65	1.62 ± 0.90	1.76 ± 0.65	1.66 ± 1.25
Placebo	0.55 ± 0.29	1.51 ± 0.50	1.84 ± 0.64	2.34 ± 1.03	2.36 ± 0.60

Not significant differences between groups through the study period.

Table IV
Correlations between changes in biochemical characteristics and changes in some anthropometric indices during follow-up

	% Δ Serum zinc	% Δ Hair zinc	% Δ Leptin	% Δ IGF-I	% Δ IGFBP-3
Δ Sum 4 skinfolds (mm)					
Supplement	0.49	0.61	0.01	-0.26	-0.61
Placebo	-0.25	-0.25	0.71**	0.38	0.07
% Δ Sum 4 skinfolds (mm)					
Supplement	0.45	0.85*	-0.04	-0.37	-0.56
Placebo	0.10	0.34	0.30	-0.10	0.06
Δ Weight/age z-score					
Supplement	0.69*	0.80*	0.20	-0.09	-0.47
Placebo	-0.45	0.45	0.50	0.58*	0.58*
% Δ Weight/age z-score					
Supplement	0.63*	0.54	0.13	-0.05	-0.43
Placebo	-0.43	0.46	0.68**	0.62*	0.49
Δ Length/age z-score					
Supplement	0.82**	0.84*	-0.07	0.22	-0.48
Placebo	-0.41	0.61	0.14	0.25	0.47
% Δ Length/age z-score					
Supplement	-0.54	-0.03	0.62	0.22	0.87**
Placebo	0.38	0.03	0.17	-0.03	-0.32

*P < 0.05. **P < 0.01.

(SPSS, Chicago, IL, USA). All the results for continuous variables were expressed as mean ± standard deviation. Mean changes were compared by using the analysis of variance test for repeated measurements. Post-hoc comparisons were done by the Tukey test. For simple comparison between two means the Student's t test was used. Changes in anthropometric and biochemical data between baseline and 6 month of follow-up were calculated as absolute values (D) and also as percentage of the baseline value (%D). Simple linear correlations are expressed with r Pearson's coefficient. $P < 0.05$ was taken as the limit of significance.

Results

Baseline data did not differ in the variables studied between supplement and placebo groups (tables I-III). Infants of both groups were comparable for male:female ratio (8/6 *vs* 8/9) and ponderal index (2.1 ± 0.2 *vs* 2.2 ± 0.4). The analysis of the type of feeding the infants received during the follow-up only showed a significant difference for the introduction of solid foods, that was earlier in the zinc groups than in the placebo one ($P = 0.021$).

There were not significant differences between groups for anthropometric measurements through the study period ($P > 0.05$) (table I). We found a significant effect of the independent variable, study group, in hair zinc concentrations (table II), but not in serum zinc concentrations. Post-hoc pair-wise comparisons for hair zinc revealed that there were significant differences between groups (supplement and placebo) at 1, 2, and 6 months of age. There were not significant differences between groups for biochemical data through the study period (table III).

Changes in serum and hair zinc concentrations from baseline to 6 months, showed significant correlations with changes in weight/age and length/age z-scores, in the supplement group (table IV). Changes in leptin serum concentrations during follow-up, showed significant correlations with changes in sum of 4 skinfolds and weight/age z-score, in the placebo group. Changes in serum IGF-I concentrations showed a significant correlation with changes in weight/age z-score in the placebo group. Changes in serum IGFBP-3 showed significant correlations with changes in length/age z-score in the supplement group, and with weight/age z-score in the placebo group (table IV). Changes in hair zinc concentration through the study period showed significant correlations with changes in leptin serum concentrations from baseline to 6 months of follow-up (table V).

Tabla V
Correlations between changes in zinc status and changes in leptin concentrations during follow-up

	% Δ Serum zinc	% Δ Hair zinc
Δ leptin		
Supplement	-0.44	0.87*
Placebo	-0.34	0.70*
% Δ leptin		
Supplement	-0.39	0.99**
Placebo	-0.42	0.92**

*P < 0.05. **P < 0.01.

Discussion

The main aim of this study was to assess if zinc supplementation improved growth during the first 6 months of life in infants with IUGR. The selected newborns had a low ponderal index, reflecting asymmetric growth retardation[19] due to malnutrition during the third trimester of gestation.[20] We have not observed a significant effect of zinc supplementation in the anthropometric and nutritional status variables studied. In infants born small for gestational age, Castillo-Durán et al.[11] have observed that weight increments were greater in the supplement (3 mg of zinc as zinc acetate) than in the placebo group; changes in z-score for 6 months were -1.28 to -0.66 in the supplement group and -1.43 to -1.47 in the placebo group. In low birth weight, full term infants, Lira et al.[10] do not showed a significant effect on weight and length gains, infants given 5 mg Zn sulphate gained more weight than infants given placebo during weeks 17-26; they do not observed any effect with 1 mg zinc supplementation. In a community-based study conducted in low birth weight infants from India, Sur et al.,[9] have observed that linear growth and weight for age showed significant differences between the supplement (5 mg of elemental zinc as zinc sulphate) and placebo groups only at the end of 1 year; no significant differences were observed at 6 months of supplementation. In preterm infants, Díaz-Gómez et al.[21] have observed that the supplement group (final content of zinc in the formula 10 mg/L) had a greater linear growth when compared with the placebo group (final content of zinc in the formula 5 mg/L). In Bangladeshi infants recruited at 4 weeks of age, Osendarp et al.[22] have observed greater weight gains in the supplement (5 mg elemental zinc as zinc acetate) than in the placebo group but only in 43 infants who were zinc deficient at baseline.

In the five randomised clinical trials of zinc supplementation during the first months of life, that are comparable to our study, results are not conclusive. In the majority of the trials the effect of zinc supplementation was not very big. Of the three studies conducted in low birth weigh infants,[9-11] one observed a clear effect on weight gain,[11] one only observed a significant weight gain from 17 to 26 weeks,[10] and the other one only observed a significant effect after 1 year of supplementation.[9] Surprisingly, in the study with greater effect, zinc supplementation was lower (3 mg zinc) than in the other two studies (5 mg zinc). We provided 3 mg elemental zinc, the adequate intake for this age group being 2 mg/day.[23] The lack of effect may be due to one or more of the following reasons: 1) the age of these infants did not allow us to detect a significant effect on growth; 2) zinc might not have the primary growth limiting nutrient for these infants.

The concentrations of zinc in plasma and hair samples are frequently used to analyse zinc nutrition; however, they are not very sensitive, especially in the case of marginal deficiencies.[24] In our study we have observed significant changes of hair zinc in the supplement group when compared to the placebo group. Our infants with IUGR do not show serum zinc concentrations indicating the presence of zinc deficiency.[22] Despite the lack of effect of zinc supplementation on growth in our study, we have observed significant correlations between zinc status and changes in weight/age and length/age z-scores, but only in the supplement group. This point out that despite of a lack of significant effect of zinc supplementation, growth of IUGR with asymmetric growth retardation is related with zinc status during the first months of life.

Changes in IGF-I and IGFBP-3 were related with changes in growth through the study period; however, not significant correlations were observed between changes in zinc status and changes in these two proteins. Zinc deficiency in rats result in impaired growth accompanied by decreased and cyclic food intake. These signs are associated with decreased plasma IGF-I. It has been observed in rats that reduced food intake precedes the decreased plasma IGF-I concentration and that IGF-I is not responsible for the decreased growth and food intake of zinc-deficient rats.[25]

Changes in leptin serum concentrations during follow-up were related with changes in sum of skinfolds and weight/age z-score, but only in the placebo group. Changes in leptin serum concentrations were significant related with changes in hair zinc changes during follow-up, in both groups. Some studies have investigated the regulation of leptin levels during zinc deficiency, and they provide useful information in both the rat[26] and humans.[14] The results of these studies are entirely consistent. Circulating leptin concentrations are reduced during zinc deficiency. This reduction appears to be due to both a decrease in the amount of body fat and a decrease in the amount of leptin produced per gram of adipose tissue.[27] In subcutaneous adipose tissue, it has been observed that zinc treatment significantly increased leptin production (42 %).[28]

In a homogeneous group of IUGR infants with asymmetrical growth retardation (third trimester malnutrition), 3 mg/day zinc supplementation do not show significant improvements in weight and length growth. However, changes in zinc status were related with changes in weight and length during the first 6 months of life. Changes in leptin serum concentrations were related with anthropometric indices of body fat accretion.

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