ORIGINAL RESEARCH PAPER

Paediatrics

A STUDY OF ZINC SUPPLEMENTATION FOR NEONATAL HYPERBILIRUBINEMIA:A RANDOMIZED CONTROLLED TRIAL

KEY WORDS: Jaundice, Neonate, Zinc compounds

Dr. Mani Shankar* M.D. (Ped.), Ph.D. (Ped.), Assistant Professor, Department of Pediatrics, Darbhanga Medical College & Hospital, Laheriasarai, Bihar.*Corresponding Author

Dr. Ashok Kumar Associate Professor, Department of Pediatrics, Darbhanga Medical College & Hospital, Laheriasarai, Bihar.

ABSTRACT

Objectives: To determine the efficacy of oral zinc for treatment of idiopathic neonatal hyperbilirubinemia in near-term and term (35-41 weeks) neonates.
Design: Randomized placebo-controlled trial.
Setting: Pediatrics Department of DMCH
Participants: Eighty newborns with idiopathic neonatal hyperbilirubinemia.
Intervention: Neonates were randomized to receive either oral zinc sulfate (10 mg/d) or placebo for 7 days.
Main outcome measures: Primary: total serum bilirubin levels at48 (±12) h, 96 (±12) h and 144 (±12) h after intervention.
Secondary: duration of phototherapy, and serum zinc and copper levels.
Results: Baseline mean (SD) total serum bilirubin levels were14.8 (3.8) and 14.4 (3.5) mg/dL in zinc and placebo groups, respectively. No significant differences were observed in total bilirubin levels between the two groups after the intervention.
Mean (SD) total serum bilirubin levels in zinc and placebo groupswere 13.9 (2.5) vs. 13.4 (1.9) mg/dL (mean difference 0.566; 95%CI - 0.535, 1.668, P=0.038) at 48 h, 13.1 (2.7) vs. 12.8 (2.3) mg/dL(mean difference 0.234; 95% CI -1.011, 1.479, P =0.708) at 96 hand 8.0 (2.0) vs. 8.6 (1.2) mg/dL (mean difference -0.569, 95% CI-1.382, 0.242, P=0.166) at 144 h. Although the mean duration of phototherapy in the zinc group was less by 21.3 h (95% CI 11.6,30.9, P=0.052), the difference was not significant. Post intervention, serum zinc levels were significantly higher in thezinc-supplemented group while serum copper levels were comparable between the two groups.
Conclusions: Oral zinc sulfate, in a dose of 10 mg/day, isnot effective in the management of idiopathic neonatal hyperbilirubinemia.

INTRODUCTION

Standard treatment for neonatal hyperbilirubinemia consists of phototherapy and/or exchange transfusion depending on serum bilirubin levels. Although effective, boththese modalities necessitate hospital admission, increase the cost of care, expose the baby to the risk of infection, and are associated with side effects [1,2]. Enterohepatic circulation (EHC) of bilirubin contributes significantly to neonatal hyperbilirubinemia [3], and its blockage might be a therapeutic target. Oral zinc salts have been shown to decrease serum bilirubin levels in hyperbilirubinemic rats, presumably by inhibition of EHC [4]. However, the role of zinc in disrupting EHC in human neonates is not clear. A recent trial has failed to demonstrate any beneficial effect of zinc on the incidence of hyperbilirubinemia in at-risk neonates [5].

This randomized, placebo-controlled trial was conducted to determine the effect of oral zinc supplementation on total serum bilirubin (TSB) levels in idiopathic neonatal hyperbilirubinemia.

METHODS

The study was conducted in Department of Pediatrics, Darbhanga Medical College & Hospital, Laheriasarai, Bihar. It was approved by the Institute Ethics Committee. Study population included term (37-41 weeks) and near-term (35-36 weeks) newborns with idiopathic neonatal hyperbilirubinemia. The inclusion criteria were: (i) appearance of jaundice after 24 hours of age; (ii) total serum bilirubin (TSB) exceeding the 40th percentile track for age as per the hour- specific bilirubin nomogram of the American Academy of Pediatrics (AAP) [6]; and (iii) absence of any obvious cause of neonatal hyperbilirubinemia. Exclusion criteria of the study were: (i) presence of any systemic illness, and (ii) major congenital malformations. A written informed consent was taken from all parents before inclusion of their child in the study.

The bottles of zinc sulfate (10 mg/mL; 10mL) and placebo were procured from Apex Pharmaceuticals Private Limited, Chennai, India. Bottles and contents for both the groups were identical in color and appearance. Information about the content of bottles was supplied in separate sealed envelope that was not opened until the end of study. Randomization was done using random numbers table. These numbers were written on the bottles, taking one bottle from each box, with each block consisting of 6 or 8.
Following investigations were done to exclude known cause of jaundice: blood group of the mother and baby, hemoglobin, complete blood count, reticulocyte count, peripheral blood smear examination, Coomb's test, free T4 and thyroid stimulating hormone (TSH), and G6PD assay. Neonates were managed as per AAP clinical practice guidelines [7]. The infants were given 0.5 mL of the solution (either zinc sulfate or placebo) - twice daily by the mother - for 7 days, using a calibrated dropper provided with the bottle. TSB levels were estimated at 48 (12) h, 96h and 144 (12) h, after starting the intervention, or more frequently, if indicated. The total duration of phototherapy was noted.

Any adverse effect like retching, vomiting, abdominal distension, diarrhea, skin rash, irritability/crying, change in infant's behavior or physical signs were noted by parents and/or physicians. Bottles were checked for residual content to ensure proper utilization. Serum zinc and copper levels were estimated using atomic absorption spectrophotometry (Perkin Elmer, USA, Model No. 2380) in infant's blood at baseline and at the end of intervention.
The primary outcome variables were TSB levels at 48 (±12) h, 96 (±12) h and 144 (±12) h after starting the intervention. Secondary outcome variables were the duration of phototherapy, and serum zinc and copper levels at the end of intervention.

With a probability of type I error ( ) of 0.05 and a power of 0.90, a minimum sample size of 37 newborns in each group was calculated to detect a reduction of at least 3 mg/dL in the mean TSB levels in the zinc supplemented group. To account for loss to follow-up, a total of 40 newborns were enrolled in each group.

Statistical analysis:

Data were entered in Microsoft excelsoftware and analyzed by SPSS version 16.0. Parametric data were expressed as percentage, mean, median and inter quartile range. Student's t-test was used to analyze continuous variables. Chi-squared test was used to test associations between all categorical variables and Fisher exact probability test was applied to compare proportions. To compare multiple means, ANOVA test was used and 95% confidence intervals (CI) were calculated. A P value <0.05 was taken as statistically significant.

RESULTS

Eighty term and near-term babies were randomly allocatedto receive zinc (n=40) or placebo (n=40). Fig. 1 shows the flow of participants in the study. Both the groups were comparable for birth weight, gestational age, proportion of low birth weight newborns, gender, mode of feeding, weight loss, age of intervention and laboratory parameters. The baseline serum bilirubin, serum zinc and copper levels were also similar between placebo and zinc groups (Table I).

After intervention, no difference in TSB levels was observed between placebo and zinc groups anytime during the study period. The duration of phototherapy was 21.3 h less in zinc group in comparison with placebo but the difference did not reach statistical significance (Table II). None of the newborns in the two groups had a TSB level exceeding 20 mg/dL and none required exchange transfusion. At the end of intervention, serum zinc levels were significantly higher in the zinc supplemented group while serum copper levels were comparable between the two groups. The adverse events were comparable between the two groups (vomiting 3 vs. 2; skin rash 3 vs. 3; diarrhea 4 vs. 3; excessive cry 2 vs. 2 in placebo and zinc group, respectively).

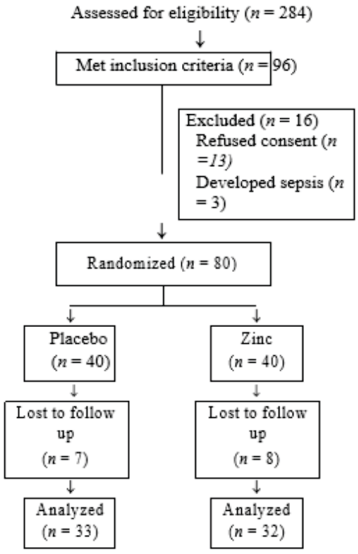

Fig. 1 Flow of participants in the study.

TABLE I BASELINECHARACTERISTICS OFSTUDYPOPULATION

Parameters	Zinc (n=40)	Placebo (n=40)
Maternal age, y (Mean±SD)	25.2 ± 4.1	26.4±3.8
History of jaundice in sibling, n(%)	4 (10)	2(5.0)
Parity, median	1	1
Cesarean delivery, n (%)	24(60.0)	20(50.0)
Gestational age (wks) (Mean ± SD)	37.6±1.5	37.7±1.4
Near terms (35-36 wks), n (%)	7(17.6)	5(12.5)
Birth weight (g) (Mean ± SD)	2711±475	2672±400
Low birth weight (<2500g), n (%)	14(35.0)	15(37.5)
Weight loss >10%, n (%)	4(10)	3(7.5)
Weight at enrolment (g) (Mean ± SD)	2595±427	2554±416
Males, n (%)	28(70.0)	25(62.5)
Exclusively breast fed, n (%)	36(90.0)	34(85.0)
Hematocrit (Mean ± SD)	48.3±5.1	47.5±4.3
Age (h) at enrolment, Median (IQR)	82.4(52,132)	78.5 (54,125)
Serum zinc levels (μg/dL)	54.8±5.9	48.4±6.5
Serum copper levels (μg/dL)	60.5±16.3	54.4±15.8
Serum bilirubin levels, mg/dL, (Mean ± SD)	13.9±2.5	13.4±1.9

TABLE II COMPARISON OFOUTCOMEVARIABLES INSTUDYPOPULATION

	Zinc (n=40)	*Placebo (n=40)*	*Mean difference (95% CI)*	*P value*
#TSB levels at 48 ± 12 h, Mean ± SD	13.9 ± 2.5	13.4 ± 1.9	0.566 (-0.535, 1.668)	0.308
#TSB levels at 96 ± 12 h, Mean ± SD	13.1 ± 2.7	12.8 ± 2.3	0.234 (-1.011, 1.479)	0.708
#TSB levels at 144 ± 12 h, Mean ± SD	8.0 ± 2.0	8.6 ± 1.2	-0.569 (-1.382, 0.242)	0.166
Phototherapy given, n (%)	21 (52.0)	18 (45.0)	0.736* (0.282, 1.921)	0.629
Age at starting phototherapy, h, Median (IQR)	87(68, 104)	92 (81, 105)	-2.46 (-16.950, 12.029)	0.733
Duration of phototherapy, h, Mean ± SD	61.9 ± 12.1	83.3 ± 17.6	-21.292 (-30.954, -11.629)	0.052
#Post-intervention serum zinc levels (μg/dL)	72.2 ± 25.7	47.8 ± 17.7	24.394 (13.475, 35.312)	0.000
#Post-intervention serum copper levels (μg/dL)	46.2 ± 16.0	48.6 ± 15.2	-2.468 (-10.214, 5.279)	0.527

TSB - Total serum bilirubin (mg/dL); * Odds ratio; # n=32 for zinc and n=33 for placebo

DISCUSSION

The present study showed that there is no beneficial effect of oral zinc in neonatal hyperbilirubinemia. TSB levels were comparable in the two groups during the intervention period. Although the mean duration of phototherapy in the zinc group was less by 21.3h, the difference was not significant. We included only idiopathic cases of neonatal hyperbilirubinemia where entero-hepatic circulation plays a dominant role in the causation of jaundice, and excluded newborns who had jaundice due to bilirubin overproduction such as ABO/Rh incompatibility, and G6PD deficiency.

The limitations of the study were small sample size and loss to follow-up. Moreover, we did not evaluate the impact of UGT1A1 gene variants on bilirubin load. In a recent study by Rana, et al. [5], the incidence ofhyperbilirubinemia was comparable in both the zinc andplacebo groups (17.9% vs19.1%). The requirement ofphototherapy was also comparable; but the duration ofphototherapy was shorter in zinc group. However, theabove study included newborns with ABO incompatibilitywhich might have influenced the results. ABOincompatibility produces hyperbilirubinemia byhemolysis, and zinc works by blocking entero-hepaticcirculation rather than preventing hemolysis.

In a study of adult patients with Gilbert syndrome, oralzinc sulfate at a dose of 100 mg daily for 7 days resulted ina significant decline in serum bilirubin levels [8]. Ourresults are at variance with this study. It may be speculatedthat zinc is ineffective in disrupting entero-hepaticcirculation of bilirubin in the face of high bilirubin loadswhich occur in newborns. There is also a possibility thatzinc may interact with intestinal flora to modulate enterohepaticcirculation of bilirubin. The relative paucity ofbacterial flora in newborns might have contributed to thelack of effect of zinc in the present study. Further, neonatalhyperbilirubinemia is typically a multifactorial disorderwith different mechanisms contributing tohyperbilirubinemia risk, and thus zinc may have a limitedimpact in this age group.

Though the duration of phototherapy was less by 21.3 h in the zinc group, the difference was not statistically significant. This could be due to small sample size as only 21 newborns in zinc group and 18 newborns in placebo group required phototherapy. This difference may be clinically relevant and further studies with a larger sample are warranted to clarify this issue. However, the present study was limited by the fact that we estimated TSB levels at fixed time intervals and this might have influenced the duration of phototherapy in present study. Future study design should include transcutaneous bilirubin estimation at more frequent intervals to detect the effect of zinc supplementation on the duration of phototherapy.

To conclude, oral zinc sulfate is not beneficial in the treatment of moderate grade idiopathic neonatal hyperbilirubinemia in term and near-term newborns.

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