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# Effect of gestational age on cord blood plasma copper, zinc, magnesium and albumin

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## Abstract

*Background*: The transport of essential trace elements from mother to fetus varies throughout gestation, and the role of transport proteins in the neonate and the mother may change during pregnancy. Magnesium, often used as tocolytic agent, may reach the fetus and appear in cord blood at higher than normal concentrations. *Aims*: To determine cord blood plasma zinc, copper and magnesium concentrations, as well as plasma albumin in premature and full-term newborns, and correlate these values with those of maternal blood plasma at birth. Also, to examine whether cord blood plasma concentration of these elements varies with gestational age. *Subjects*: The 35 mother–infant pairs included: 11 in the 38–42-week gestational age (GA), 9 in the 34–37-week GA, 11 in the 29–33-week GA group and 4 in the 24–28-week GA. Magnesium for tocolysis was given to five of the mothers in the 29–33-week GA cohort and two of the women giving birth at 24–28-week GA. *Results*: Trend analysis showed that while cord plasma zinc decreased with GA at birth, the reverse was observed for copper. There were no differences with GA either in maternal plasma zinc or copper. However, maternal ceruloplasmin tended to decrease with GA ($P = 0.0174$). Maternal and cord blood plasma magnesium exhibited a strong correlation ($r = 0.942$, $P < 0.001$), as well as between cord plasma magnesium and zinc ($r = 0.448$, $P < 0.01$). *Conclusions*: While the vigorous mother-to-fetus uphill zinc transfer is clear throughout the last trimester, copper remains in cord blood plasma at much lower concentrations than in the mother, suggesting that prematurity may

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place the newborn infant at a greater risk than the term infant to copper deficiency. This situation, together with a reduced synthesis in the fetus of the transport protein ceruloplasmin, creates another potential challenge in the nutritional support of the premature infant.

*Keywords:* Gestational age; Cord blood plasma zinc, copper and magnesium concentrations; Infants

## 1. Introduction

The role of trace elements in fetal development and growth is well documented in the literature [1,2]. Copper is of particular importance, especially early in life, for the development and maintenance of myelin [3–5]; zinc is essential for normal embryogenesis, fetal growth and protein synthesis [6–8]; magnesium is critical for bone formation, cellular integrity and multiple enzyme functions [9]. In cases of preterm labor, the fetus may be exposed to relatively large amounts of magnesium because it is used extensively as a tocolytic agent [10,11]. Deficiency of trace elements during intrauterine existence is closely related to mortality and morbidity in the newborn [1,4,6,7].

In many membrane transport processes, such as intestinal absorption or placental transfer, many divalent cations compete with each other for the same mechanisms [12,13]. It has been reported that the concentration of copper in cord blood is lower than that in mothers' blood, and that copper concentration of premature infants' cord blood is higher than that of full-term newborns [14–16]. Conversely, cord blood zinc is higher than in maternal blood plasma and decreases with gestational age (GA) [5,15,17]. However, it is not known if there is a relationship between zinc and copper transport across the placenta. Since the transport of several essential divalent elements across membranes is competitive, premature infants may have accumulated proportionately less copper than full-term infants [18] because the transport of essential trace elements from mother to fetus may vary at different stages of gestation. How much of the pharmacological doses of magnesium used as a tocolytic agent reach the fetus and appear in cord blood has not been clearly established. Similarly, in pre-eclampsia and eclampsia, which are treated with magnesium as well, the transport of trace elements from mother to fetus may be altered. There is also limited information on how pregnancy-related complications may affect the placental transfer process [6,14,19–21].

Carrier proteins are essential for the transport of these elements from the maternal circulation to the placenta [14,22]. The fetus is dependent on its own synthesis of these transport proteins for delivery of minerals to all organs of its body, but it is not well known to what extent these carrier proteins are present late in gestation and at birth. Since the absolute amount of nutrients, vitamins and trace elements transported across the placenta increases as gestation progresses, potential nutritional deficiencies may be critical in premature fetuses if the transfer of essential elements from mother to fetus has not kept pace with the needs of the fetus. Such insufficiencies may result in a reduced accumulation of these elements in storage organs of the fetus such as liver or bone. Therefore, the neonatologist is faced with the challenge of providing adequate nutrition in the early neonatal period, especially to the premature infant, whose possible specific mineral deficiencies at birth are not well determined.

The purpose of this study was to determine cord blood plasma zinc, copper and magnesium concentrations, as well as ceruloplasmin activity and albumin in premature and full-term newborns, and correlate these values with those in maternal blood plasma at birth. Another purpose is to ascertain whether cord blood plasma concentration of these elements varies with gestational age and birth weight.

## 2. Patients and methods

Cord blood samples were collected from newborns delivered at North Shore University Hospital at Manhasset over a period of approximately 6 months. Pregnant women in labor were requested to participate in this study and provide a 5-ml sample of venous blood, as well as authorizing to obtain approximately 5 ml of cord blood at delivery. Only mothers with singleton live born infants without any visible congenital abnormalities were included in the study. Both appropriate for gestational age (AGA) and small for gestational age (SGA) infants were included. Specimens were collected in a metal-free (blue top) heparinized tube. All blood samples were centrifuged at $600 \times g$ for 15 min and plasma was separated for analysis. If needed, plasma specimens were kept frozen at $-20$ °C until analysis, but not in excess of 30 days. This study was approved by the Institutional Review Board.

Copper, zinc and magnesium were determined by atomic absorption spectrophotometry (Varian SpectrAA-10, Sunnyvale, CA) using external standards. Ceruloplasmin was assayed by a spectrophotometric method based on the oxidative capacity of this protein [23]. Albumin was determined by a bromcresol green binding method (Sigma 631-2). Data were analyzed by ANOVA and Tukey's test. If data were not normally distributed, the Kruskal–Wallis nonparametric test was applied [24]. Operationally, Sigmastat® (SPSS Science, Chicago, IL) was used for determining Spearman's rank order correlations, and the correlation coefficients were obtained from the equations given by Sigmaplot® (SPSS Science). A Wilcoxon-type test for trends [25] was applied using an ad hoc computer program (Arcus®, I.E. Buchan, Liverpool, UK). The threshold of significance was 0.05.

## 3. Results

There were 11 full-term infants in the study (GA: 38–42 weeks). The male/female (M/F) ratio was 7:4 and their combined average weight (± S.D.) was 3498 ± 477 g. The nine newborns in the 34–37-week GA cohort had a M/F ratio of 4:5 and a mean weight of 2551 ± 516 g. There were 11 infants in the 29–33-week GA group (M/F: 7:4) with a mean birth weight of 1821 ± 535 g. The four 24–28-week GA premature infants (M/F: 2:2) had a mean birth weight of 785 ± 158 g. None of the mothers of the 38–42- and 34–37-week GA infants received magnesium treatment during labor.

Table 1
Maternal and cord plasma zinc and magnesium according to gestational ages

| GA (weeks) | *N* | Maternal plasma zinc (μg/dl) | Cord plasma zinc (μg/dl) | Maternal/cord zinc ratio | Maternal plasma magnesium (mg/dl) | Cord plasma magnesium (mg/dl) | Maternal/cord magnesium ratio |
|---|---|---|---|---|---|---|---|
| 38–42 | 11 | 62.5 ± 18.3 | 87.0 ± 9.5 | 0.72 ± 0.21 | 2.6 ± 0.8 | 2.8 ± 0.8 | 0.93 ± 0.13 |
| 34–37 | 9 | 52.2 ± 10.6 | 89.4 ± 14.9 | 0.59 ± 0.10 | 2.4 ± 1.5 | 2.6 ± 1.5 | 0.89 ± 0.18 |
| 29–33 | 11 | 58.6 ± 8.4 | 94.1 ± 18.7 | 0.65 ± 0.19 | 4.2 ± 3.1 | 4.7 ± 3.5 | 0.94 ± 0.18 |
| 24–28 | 4 | 55.0 ± 10.8 | 116.3 ± 45.2 | 0.51 ± 0.14 | 4.2 ± 1.2 | 3.7 ± 2.1 | 1.33 ± 0.59 |

Data are means ± S.D.

A total of 5 out of 11 in the 29–33-week GA cohort were given magnesium tocolysis and 2 of the 4 mothers giving birth at 24–28-week GA received this treatment.

Maternal and cord plasma zinc data are tabulated in Table 1. While there were no discernible differences for zinc according to GA, the maternal/cord plasma zinc ratio increased with GA ($P < 0.0001$) [25]. In addition, we found that maternal plasma albumin correlated with the corresponding zinc concentration ($r = 0.540$, $P < 0.001$, Fig. 1). Maternal and cord plasma magnesium showed a significant increment in mothers and infants of the two earliest GA groups, in association with about half of them having received magnesium for tocolysis. The relationship between this treatment and cord plasma magnesium was substantiated by the uniformity of the maternal/cord plasma magnesium ratio (Table 1), as well as the very significant correlation of magnesium values

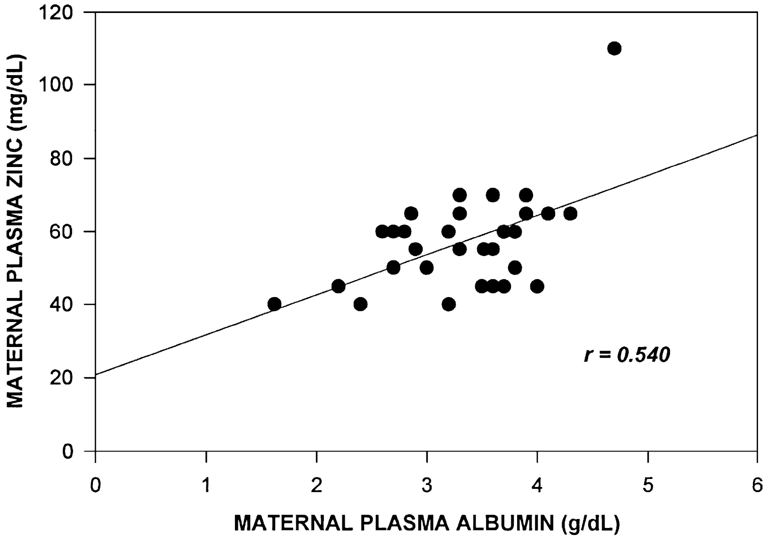

Fig. 1. The relationship between maternal plasma albumin and zinc concentrations showed a very significant positive correlation ($P < 0.001$).

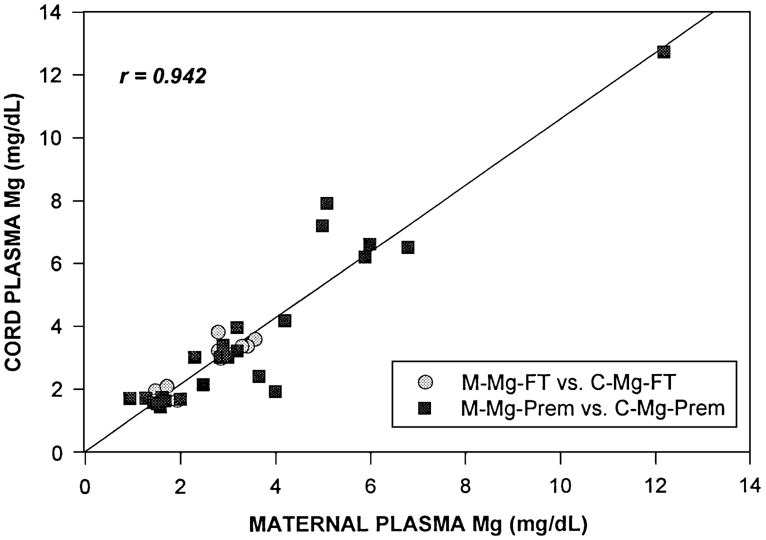

Fig. 2. The correlation between maternal (M) and cord (C) plasma magnesium was very significant ($P < 0.001$) for all groups of infants taken together. Symbols for Mg-FT correspond to the 38–42-week GA cohort; the remaining symbols (Prem) include M and C Mg of the 34–37, the 29–33, and the 24–28-week GA groups.

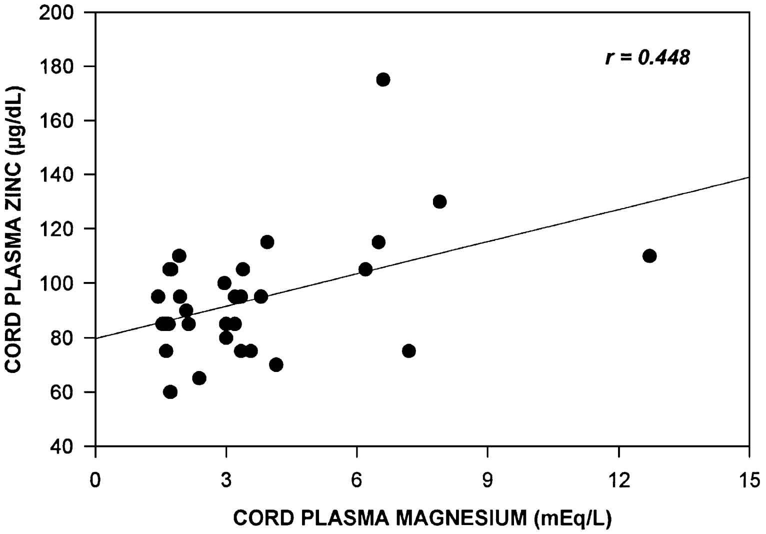

Fig. 3. Regression between cord plasma magnesium and zinc. This relationship is significant ($P < 0.01$).

Table 2
Maternal and cord plasma copper and ceruloplasmin according to gestational ages

| GA (weeks) | *N* | Maternal plasma copper (μg/dl) | Cord plasma copper (μg/dl) | Maternal/cord copper ratio | Maternal plasma ceruloplasmin (mg/l) | Cord plasma ceruloplasmin (mg/l) |
|---|---|---|---|---|---|---|
| 38–42 | 11 | 171.5 ± 26.8 | 36.0 ± 7.7 | 4.91 ± 1.05 | 291.6 ± 52.1 | 6.2 ± 4.7 |
| 34–37 | 9 | 162.1 ± 39.0 | 25.7 ± 13.9 | 7.79 ± 3.85 | 276.8 ± 63.8 | 3.9 ± 8.4 |
| 29–33 | 11 | 190.9 ± 24.5 | 39.2 ± 21.7 | 5.51 ± 2.23 | 328.0 ± 100.9 | 11.3 ± 22.1 |
| 24–28 | 4 | 193.1 ± 29.2 | 27.1 ± 13.7 | 8.39 ± 3.43 | 407.0 ± 67.0 | 9.0 ± 11.3 |

Data are means ± S.D.

between the two sampling compartments (Fig. 2). Interestingly, it appeared to be a direct correlation between cord plasma magnesium and cord plasma zinc ($r = 0.448$, $P < 0.01$, Fig. 3).

Table 2 contains data of maternal and cord plasma copper and ceruloplasmin. Although no significant correlation could be established between cord plasma zinc and copper, trend analysis revealed that zinc values decreased with GA ($P < 0.0001$), while copper did not exhibit a definite trend. No significant correlation was found between corresponding values of cord plasma zinc and copper. However, the trend of change in reverse sense between cord plasma zinc and copper was very significant ($P < 0.0001$). Maternal plasma copper showed no differences due to newborn GA at delivery. A similar uniformity was found for cord plasma copper and ceruloplasmin, with no discernible trend for either one. Maternal ceruloplasmin, in contrast, decreased with GA ($P = 0.0174$) [25]. As expected, maternal plasma copper is highly correlated with ceruloplasmin ($P < 0.0001$, Fig. 4).

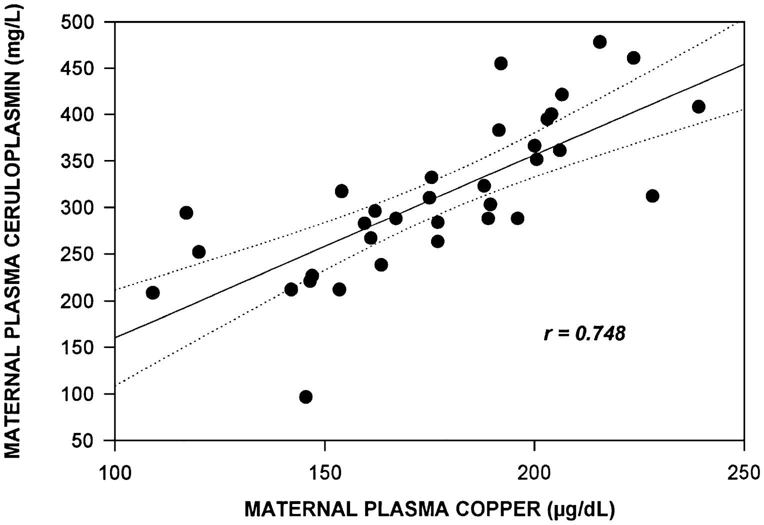

Fig. 4. Correlation between maternal plasma copper and maternal plasma ceruloplasmin. This correlation is very significant ($P < 0.0001$). The dotted bands represent the 95% confidence intervals of the regression line.

## 4. Discussion

Although fetal nutrition has traditionally focused on the study of energy sources for the fetus and placental transport of macronutrients [26], the role of micronutrients is equally vital for cell growth and differentiation [27]. An additional important factor is the complex relationship between maternal and fetal metal transport proteins and fetal mineral sufficiency. In this study, we found that maternal plasma albumin correlated with maternal plasma zinc (Fig. 1), a finding consistent with the role that albumin may have in transporting and presenting zinc to the syncytiotrophoblast [22,28]. In addition, the concentration ratio between circulating zinc in the mother and the infant tended to decrease as GA increased (Table 1), indicating that the uphill gradient characteristic of the zinc transport during gestation [15,17,29] is more marked earlier in the last trimester, than late. Conversely, the maternal/cord copper ratio decreased with GA (Table 2). The lack of correlation between cord plasma zinc and copper does not allow to assert that in each individual case, at delivery, an increased concentration of zinc may depress the concentration of copper present, as could be expected from the well-known antagonism between the two elements [12,13]. Nevertheless, the developmental trends in placental transport of zinc and copper were clearly manifest. The results presented here for these two elements in the plasma of mothers and cord blood plasma of term infants are comparable to those reported earlier [16,29,30].

It also appears as if the administration of magnesium to the mother for tocolysis permeates the placenta and essentially equilibrates with fetal circulation, denoting a diffusion phenomenon (Fig. 2). The transient neonatal hypermagnesemia, following maternal magnesium treatment, is well documented [11]. Administration of magnesium during labor appears to have an additional consequence in relation to zinc placental transport. The data obtained show an association between cord plasma magnesium and zinc. It is not clear whether a surge in zinc diffusion attributable to magnesium administration, or other membrane phenomena, are involved in this process.

The known hypercupremia of gestation [5,14,15,17] appears to be more marked in those women who delivered prematurely (Table 2). Concurrently, maternal ceruloplasmin tends to decrease as GA increases. This is consistent with the downhill gradient of plasma copper from mother to infant, which is more marked in 24–28-week GA newborns than in the more mature infants. The results we obtained show that cord plasma ceruloplasmin concentration is much lower than the respective maternal values. A previous report in the literature also showed a several-fold difference between maternal and cord blood plasma copper [31]. However, the methodology used more than three decades ago and the absolute values reported elicit questions regarding accuracy. The parsimonious delivery of copper to the fetus is consistent with its inability to synthesize greater amounts of ceruloplasmin, or, alternatively, a physiological consequence of the greater zinc influx into the fetal compartment, which inhibits copper translocation by an increase of placental metallothionein. This possibility is supported by the finding of lower concentrations of copper in the liver of fetuses of rats fed with high zinc diets than in normally fed controls [32]. The test for ceruloplasmin used in this study is dependent on the so-called ferrooxidase capacity of ceruloplasmin, which may not be equivalent to the total amount of ceruloplasmin that could be revealed by immunoreactivity. In pregnancy, it appears as if maternal plasma ceruloplasmin to copper ratio is highest in the youngest premature infants, simultaneously with the steepest maternal to cord plasma copper ratio. This may be indicative of the importance of maternal ceruloplasmin during gestation as the key copper carrier for placental transport, and clearly apparent from Fig. 4.

Only limited data have been reported on possible relationships between GA and trace element concentration in cord blood plasma. The decreasing trend of zinc concentrations with increased GA previously published [15] was less marked in the present population. In the case of copper, while maternal plasma values were similar, cord blood plasma copper concentrations were significantly lower in the data presented here than in those obtained in Poland, but closer to the values obtained in Indian women and infants [5].

All mechanisms for the placental transfer of divalent cations in humans have not been fully elucidated. Low atomic weight elements such as magnesium appear to cross the placenta entirely by diffusion. However, heavier elements, such as zinc and copper, show requirements for transport proteins or transmembrane carriers. On the maternal side, these molecular vehicles are partially dependent on the mothers' nutritional status, and, on the fetal side, on the timely synthesis of the respective transporters for distribution throughout the body. Prematurity presents a challenge to the neonatologist in the need to compensate the interrupted normal accretion of essential elements by nutritional means after birth. Other factors, including multiple gestations and their related problems, now increasingly frequent, may further distort the delicate intrauterine nutritional buildup.

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