Serum Concentrations of 10 Acute-Phase Proteins in Healthy Term and Preterm Infants from Birth to Age 6 Months

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Aiming to define the evolution pattern of 10 acute-phase proteins in early infancy, we measured nephelometrically the serum concentrations of albumin, prealbumin, retinol-binding protein, transferrin, ceruloplasmin, hemopexin, haptoglobin, α1-acid glycoprotein, α2-macroglobulin, and α1-antitrypsin in 395 term and preterm infants (gestational ages 26–41 weeks). Measurements were performed within 24 h after birth and then at the end of 1 (n = 171), 3 (n = 155), and 6 (n = 90) months afterwards. Data obtained from 250 healthy adults were used as adult reference values. All proteins increased progressively with postnatal age, except for α1-antitrypsin, which remained stable from birth to the 6th month. Concentrations of almost all measured proteins were significantly lower in preterm than in term infants in the first 3 months. Compared with adult values, α2-macroglobulin and α1-antitrypsin were higher in infants throughout the 6 months. The other proteins were significantly lower at birth than adult values but after 6 months, only albumin, prealbumin, retinol-binding protein, and α1-acid glycoprotein still remained lower in infants. Thus both gestational and postnatal age should be considered when interpreting concentrations of these proteins in early infancy.

Indexing Terms: nephelometry/α1-acid glycoprotein/α2-macroglobulin/α1-antitrypsin/albumin/prealbumin/ceruloplasmin/hemopexin/haptoglobin/retinol-binding protein/transferrin

Acute-phase proteins in infancy are of special interest because changes in their concentrations in serum are helpful for diagnosing and following up the course of systemic infections (1–4), for assessing intrauterine and postnatal nutritional status (5–8), and as predictors of the development of respiratory distress syndrome and bronchopulmonary dysplasia (9, 10). Reference values of acute-phase proteins in infancy, as presented by most studies to date, refer to individual proteins such as albumin (11–13), prealbumin (6, 14), α1-acid glycoprotein (2, 4, 13, 15), and ceruloplasmin (1, 3, 16), as measured by various methods. Moreover, data on the changes in concentrations of acute-phase proteins during infancy are limited: α1-acid glycoprotein during the first month postpartum (15), albumin during the first 8 weeks (11), and transferrin and ceruloplasmin during the first year (16, 17).

Here, we have utilized nephelometry to sequentially measure serum concentrations of 10 acute-phase proteins in preterm and term infants during the first 6 months postpartum in an attempt to define the changes in concentrations of these proteins in early infancy.

Materials and Methods

Three hundred ninety-five infants appropriate for gestational age (gestational age 26 to 41 weeks), 221 boys and 174 girls, were studied. The infants were classified into two groups: term infants, 242 infants with gestational ages of 38–41 (39.5 ± 1) weeks and birth weight 3387 ± 331 g, and preterm infants, 153 infants with gestational ages of 26–37 (33 ± 3) weeks and birth weight 1958 ± 606 g. Written informed consent was obtained from all parents. After detailed maternal and perinatal history was recorded, neonates meeting the following criteria were entered in the study: (a) known gestational age, as determined by prenatal ultrasonography; (b) Apgar score > 7 at 5 min; (c) no evidence of severe respiratory distress syndrome or perinatal asphyxia; (d) no major congenital malformations; and (e) no evidence of intrauterine or perinatal infections.

Within 24 h after birth, 0.5 mL of whole blood was collected from all neonates, through an indwelling umbilical arterial catheter or by venipuncture, during routine procedures. Subsequent samples were taken at the end of ages 1, 3, and 6 months. Before collecting blood, we clinically examined the infants and collected data for conditions that could possibly affect serum concentrations of acute-phase proteins, such as infections, malnutrition, recent surgery, and renal or liver disease. Infants who had undergone double-volume exchange transfusion or had a history of the above-mentioned disease states were excluded from follow-up.

Measurements performed in 250 adults, 116 men (ages 36 ± 8 years) and 134 women (ages 38 ± 6.7 years), who were known, apparently healthy, volunteer blood donors, were used to determine adult reference values.

Serum specimens, separated by centrifugation at 200g, were stored at −70°C until analysis. Protein measurements were performed by nephelometry with the Behring Nephelometer Analyzer (BNA, Behringwerke AG, Marburg, Germany). Reference curves were prepared by using the Behring N Protein Standard

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Serum for quantitative immunochemical determinations of albumin, prealbumin, transferrin, α1-acid glycoprotein, α2-macroglobulin, haptoglobin, hemopexin, ceruloplasmin, and retinol-binding protein; the N Protein Standard PY (Code OUID) was used for α1-antitrypsin (both N proteins approved by the Paul Ehrlich Institute, Federal Office for Sera and Vaccines, Lanen, Germany), and their values were assigned with reference to the reference values of the International Federation of Clinical Chemistry (IFCC). The antisera used for quantitative immunochemical determination of serum concentrations of the above proteins were NA Reagents (Behringwerke), also approved by the Paul Ehrlich Institute, Federal Office for Sera and Vaccines. For accuracy and precision of the given results, we established our own control values and confidence limits.

For statistical analysis, we calculated the 5th, 50th, and 95th percentiles of serum concentrations for each group of infants at each postnatal age. Curves of the 5th and 95th percentiles of each protein, plotted against postnatal age, were constructed for both groups of infants in comparison with the adult reference range. The distribution of values was not the same for all proteins at all ages in the examined groups. On the first day after birth, the distribution of values of most proteins was not gaussian in either group of infants. On follow-up, values for all proteins showed a normal distribution in term infants, whereas in the preterm infants retinol-binding protein and ceruloplasmin at the first month and retinol-binding protein and haptoglobin at the third month did not exhibit normal distribution. In adults, most of the proteins except for albumin and prealbumin showed a normal distribution. Therefore, we used a nonparametric method (Mann–Whitney U–Wilcoxon rank sum W test) to compare the significance of the differences between the groups (18). Data were analyzed with the use of the SPSS for MS Windows, Release 6.1 (License: 814525, International Use) software package.

Results and Discussion

The 5th, 50th, and 95th percentiles of serum concentrations of the 10 acute-phase proteins in the two groups of infants and in adults are shown in Table 1. In addition, Fig. 1 shows the pattern of changes in concentrations for four of the proteins (prealbumin, transferrin, hemopexin, and α2-macroglobulin), as examples. The patterns of change for the 10 proteins were similar in term and preterm infants. Concentrations increased progressively from birth to the 6th month, except for α1-antitrypsin, which remained more or less stable during this period. With few exceptions, the concentrations of these proteins were significantly lower in preterm than in term infants during the first 3 months postpartum. By 6 months, only ceruloplasmin was still significantly lower in preterm infants (Table 1). Lower concentrations of acute-phase proteins in preterm neonates than in term neonates have been previously reported (2, 14, 15, 19, 20); however, our results indicate that gestational age significantly affects the serum concentrations of these proteins not only at birth but also during the next 3 months.

Considering the values reported by other researchers using nephelometry, ours seem very close to those reported by Raubenstein et al. (8) in a group of 21 neonates, but the α1-acid glycoprotein values measured at birth were higher than those reported by Sann et al. (15): 0.18 ± 0.08 and 0.15 ± 0.09 g/L in term and preterm neonates, respectively.

Haptoglobin concentrations at birth could not be assessed, being below the detection limit of the nephelometer (<0.112 g/L) in 80% of the preterm infants and in 60% of the term infants. During the following months, the percentage of infants with measurable haptoglobin increased progressively; by the sixth month, it could be measured in all infants. These findings confirm the previous reports about haptoglobin concentrations being undetectable in neonates (13).

Comparison between the values for term infants and for adults showed that α2-macroglobulin and α1-antitrypsin concentrations were significantly higher in infants throughout the period studied. High amounts of protease inhibitors in neonates have been previously reported by Levine et al. (21, 22), who suggested that this increase may reflect a response to excessive protease uptake across the immature intestinal barrier of the neonate (21). Our results not only confirm these findings in a larger group of neonates but further extend this observation beyond the neonatal period.

Ceruloplasmin and transferrin concentrations at birth were lower in infants than in adults. These concentrations rapidly increased in the ensuing months, exceeding adult values at 3 and 6 months, respectively (Fig. 1). The rest of the measured proteins, i.e., albumin, prealbumin (Fig. 1), retinol-binding protein, and α1-acid glycoprotein were significantly lower in infants than in adults throughout the period. The statistical significance of the difference in values between adults and term infants is shown in Table 1.

Similar results were obtained for α2-macroglobulin, α1-antitrypsin, transferrin, and ceruloplasmin concentrations between preterm infants and adults. The rest of the measured proteins remained significantly lower in the preterm infants than in adults (P < 0.05), even after 6 months (Table 1).

We conclude that adult reference ranges are not useful as reference values for infants. Furthermore, the emerging relationship between acute-phase protein concentrations and both gestational and postnatal age suggests that both ages should be considered when interpreting concentrations of these proteins during the first months after birth. Values presented in this study could be useful for clinical interpretation of such proteins in early infancy.
Table 1. Percentiles of serum concentrations (g/L) of 10 acute-phase proteins in term and preterm infants at different postnatal ages and in adults.

<table>
<thead>
<tr>
<th>Age</th>
<th>Albumin</th>
<th>Prealbumin</th>
<th>Retinol-binding protein</th>
<th>Transferrin</th>
<th>Ceruloplasmin</th>
<th>Hemopexin</th>
<th>Haptoglobin</th>
<th>α₁-Acid glycoprotein</th>
<th>α₂-Macroglobulin</th>
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<td>42 0 6.7 10 0 18 4 0 011 0 017 0 030 1 18 1 87 2 80 0 04 0 08 0 23 0 06 0 21 0 49</td>
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</table>

a Significance of the difference between term and preterm infants.

b Significance of the difference between term infants and adults (Mann-Whitney U-Willcoxon rank sum W test); NS, not significant.

c Haptoglobin was undetectable by nephelometry in 80% of the preterm and 60% of the term infants at birth.

d All differences between term and preterm infants were not significant, except for ceruloplasmin (P < 0.05).
Fig. 1. The 5th (—) and 95th (- - -) percentiles of serum concentrations of prealbumin, transferrin, hemopexin, and α2-macroglobulin in term (∅) and preterm (○) infants by postnatal age, in comparison to adult reference range (no symbols on lines).

References