Placental Transfer of Immunoglobulin G Subclasses

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The concentrations in cord blood of total immunoglobulin G (IgG) and the four subclasses of IgG were measured in 34 fetuses at a mean gestational age of 25 weeks (range, 18 to 35 weeks). The blood samples were obtained by percutaneous umbilical blood sampling, and results were compared with the respective IgG subclass concentrations of the mothers. The efficiency of transplacental transfer of the different IgG subclasses was determined. Transfer of IgG1 and IgG4 was found to be significantly more efficient than that of IgG3 and IgG2. IgG2 was the subclass least efficiently transferred from mother to fetus. These differences may partly explain the susceptibility of newborns to various pathogens, such as streptococcus group B.

Placental transfer of immunoglobulin G (IgG) has been demonstrated by several investigators (4–6, 8–10, 13, 14). It was found that the IgG concentration in the fetus correlates positively with gestational age, and the cord blood IgG level at term tends to exceed the respective maternal concentration, suggesting active placental transport from mother to fetus (5, 6, 14). However, conflicting results regarding the relative transport of the different IgG subclasses have been reported. Although some authors reported reduced placental transfer of IgG2 in comparison with that of the other subclasses (4, 13), others were unable to confirm these observations and reported similar efficiencies of passage of all four IgG subclasses (8–10).

Most investigations of placental IgG subclass transfer at different stages of pregnancy were performed by cord blood examination at the time of delivery in full-term or preterm infants. Using the new technique of percutaneous umbilical blood sampling, we obtained fetal blood during the second and third trimesters of pregnancy in order to investigate the passage of IgG subclasses through the placenta at earlier stages of intrauterine life. Our report is also novel in the sense that the infants studied were not prematurely born.

MATERIALS AND METHODS

Blood from 34 fetuses was obtained by the percutaneous umbilical blood sampling technique as previously described (7). The pregnant woman was prepared as for amniocentesis. A real-time ultrasound sector or linear scanner was used to locate the umbilical cord insertion site on the placenta. A 25-gauge needle was inserted with ultrasound guidance into the umbilical vein, and fetal blood was drawn into a syringe. The purity of the sample was assayed by Kleihauer-Betke smear and by measurement of the size of the erythrocytes. Gestational age was determined on the basis of the last reported menstrual cycle and confirmed by ultrasonographic

measurements. Peripheral venous blood was drawn from the mother at the time of the procedure.

The medical indication for the procedure was risk of fetal hemolytic disease (mainly Rh hemolytic disease) in 22 fetuses and suspected genetic disease in 12. Outcome was good in 30 fetuses; of the remaining 4, 1 fetus examined at 23 weeks' gestation had severe intrauterine growth retardation and oligohydramnios and died in utero during the third trimester and the other 3 had severe hydrops fetalis and died after delivery.

Concentrations of maternal and fetal IgG subclasses 1, 2, 3, and 4 in serum were determined by an immunoradiometric assay. Tubes were coated with mouse anti-human IgG1, IgG2, IgG3, or IgG4 specific monoclonal antibodies (clones JLS12 [IgG1], HP6014 [IgG2], 2G4 [IgG3], and RJ4 [IgG4]) purified from mouse ascites fluid. Diluted serum specimens were added and incubated for 1 h. Afterward, the tubes were washed and goat anti-human IgG coupled with ^{125}I was added and incubated for 1 h. After being washed again, the tubes were counted. Patient values, reported as milligrams per deciliter, were extrapolated from a standard curve calibrated against World Health Organization standards for each of the IgG subclasses. Three control serum samples (high, medium, and low IgG concentrations) from previously tested patient specimens were included in each run for each of the IgG subclasses. Total IgG was measured by nephelometry with the Behring system. The sum of the IgG subclasses was compared with the total IgG. If results differed by >10%, the tests were repeated.

Relative IgG subclass concentrations were calculated as percentages of total IgG; means and standard errors were also calculated. The mother-to-fetus ratios for concentrations of the different subclasses were plotted against gestational age, and linear regressions were performed on the resulting scatter diagrams. Differences in placental transfer of IgG subclasses were calculated and analyzed by the paired t test.

RESULTS

Because no differences for any of the subclasses were noted when serum IgG concentrations of the fetuses with suspected hemolytic disease were compared with those of fetuses with

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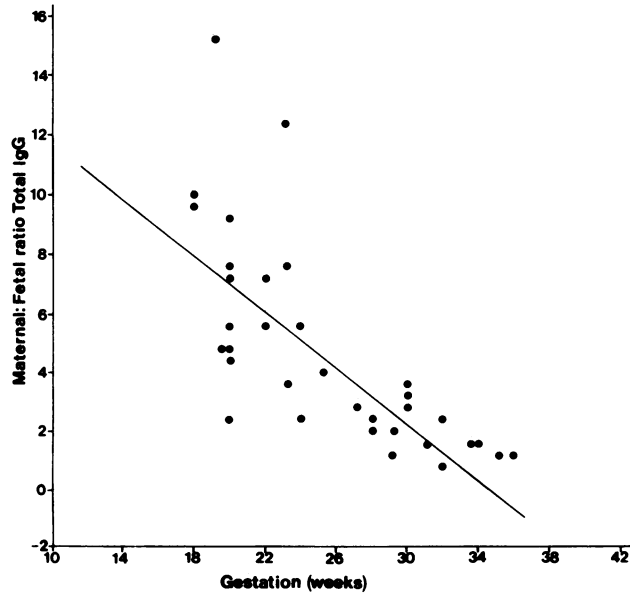
FIG. 1. Mother-to-fetus ratio of total IgG related to gestational age ($y = -0.46x + 16.4$; $r = 0.73$; $P < 0.0001$, $n = 34$).

FIG. 3. Mother-to-fetus ratio of IgG2 related to gestational age ($y = -1.02x + 34.3$; $r = 0.71$; $P < 0.0001$; $n = 34$).

genetic disorders, all 34 fetuses (and mothers) were treated as a single group. Results are based on the collective data.

Placental transfer with respect to gestational age. Figures 1 to 5 show the relationships between the placental transfer of total IgG, IgG1, IgG2, IgG3, and IgG4 (expressed as mother-to-fetus ratio) and gestational age. A linear increase in placental transfer of total IgG and IgG subclasses was found to correlate with the length of gestation. The linear regression had highly significant correlation coefficients for IgG1, IgG2, IgG3, IgG4, and total IgG ($r = 0.73, 0.71, 0.65, 0.63,$ and 0.73, respectively; $P < 0.0001$ for each).

Differences among subclasses in placental transfer. Placental transfer of IgG1 (mean ± standard error, 4.02 ± 0.50) was significantly more efficient than the transfer of IgG2 ($8.41 \pm$
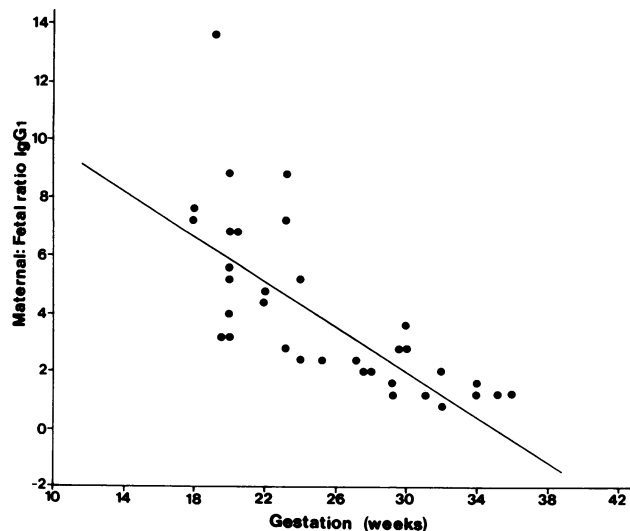
$1.35, P < 0.0001$) and IgG3 ($6.54 \pm 0.91, P < 0.001$) but similar to the transfer of IgG4 ($4.50 \pm 0.55, P < 0.1$). Transfer of IgG2 was significantly less efficient than that of IgG3 ($P < 0.04$) and IgG4 ($P < 0.001$). The difference in transport between IgG3 and IgG4 was also statistically significant ($P < 0.002$). Thus, the transfer of IgG1 and IgG4 was more efficient than the transfer of IgG3, and the transfer of IgG2 was least efficient.

DISCUSSION

Our results confirm a close linear relationship between placental transfer of each IgG subclass and gestational age, as previously shown (4, 8–10, 13). Our results also demonstrate that although all IgG subclasses cross the human placenta, their transport is not uniform: IgG1 and IgG4 are transported more efficiently than IgG3 and IgG2, and IgG2 is transported least efficiently. Reduced passage of IgG2 from mother to fetus was reported by Wang et al. (13) and Hay et al. (4), whereas Pitcher-Wilmott et al. (10) and Morell et al. (8, 9) reported similar passages for all four subclasses. There is no satisfactory explanation for these discrepant results.

FIG. 2. Mother-to-fetus ratio of IgG1 related to gestational age ($y = -0.38x + 13.8$; $r = 0.73$; $P < 0.0001$, $n = 34$).

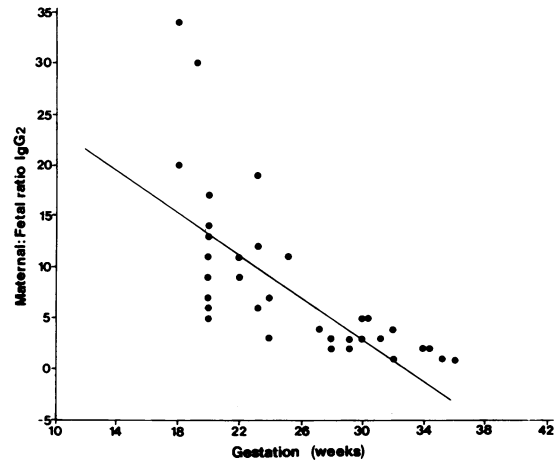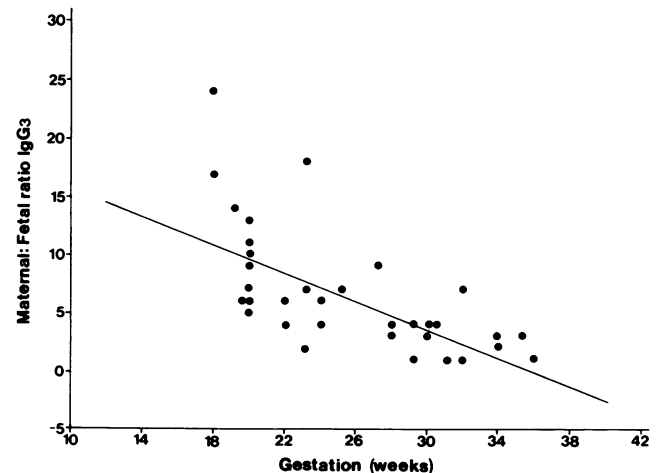FIG. 4. Mother-to-fetus ratio of IgG3 related to gestational age ($y = -0.63x + 22.5$; $r = 0.65$; $P < 0.0001$, $n = 34$).
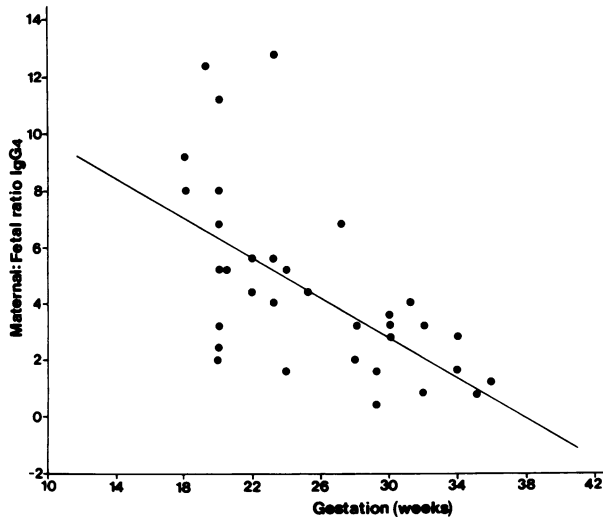
FIG. 5. Mother-to-fetus ratio of IgG4 related to gestational age ($y = -0.37x + 13.8$; $r = 0.63$, $P < 0.0001$, $n = 34$).

Fetal IgG subclass concentrations approximate maternal concentrations at 38 weeks and sometimes continue to increase to concentrations higher than maternal concentrations at delivery. The full-term infant therefore has an adequate level of IgG of all four subclasses, providing it with passive immunity to a wide range of potential pathogens. However, the lower IgG concentrations in preterm infants could account in part for their increased susceptibility to infections. In addition, the less efficient transfer of IgG2 and IgG3 could play a role in the susceptibility of the preterm and even full-term newborn to certain infections. For example, antibodies to group B streptococci are mainly IgG2, and passage of these antibodies from mother to child is less efficient (12).

In an earlier study, intravenous immunoglobulin infusion to pregnant women during the last trimester resulted in the apparent passage of all four IgG subclasses to the fetus if the delivery was after 32 weeks' gestation (11). However, only a few babies were investigated, and the efficiency of the passage of different IgG subclasses was not determined. The transfer of IgG antibodies is mediated by an active transport mechanism and appears to be initiated by receptors at the syncytiotrophoblast membrane that binds the Fc portion of the IgG molecules (11). Testing the placental transport of specific antiviral immunoglobulins revealed that IgG-specific antibodies for rubella and for cytomegalovirus are transferred more efficiently than IgG-specific antibodies for herpes simplex virus or Epstein-Barr virus (3). Also, placental transfer of antibody to polioviruses is not uniform. Gelfand et al. (2) found that type 3 poliovirus antibody was transmitted less efficiently than antibody for types 1 and 2. Similarly, it was shown that mother-to-fetus ratios for some viral antibodies are higher than others (1). This selectivity of placental transfer of antibodies to different viruses is not yet understood. The present findings of differences in transport of different IgG subclasses might help explain these findings, because it is known that the antibodies to various pathogens comprise several IgG subclasses in various proportions (12).

Of the five major classes of antibodies (IgG, IgM, IgA, IgD, and IgE), only IgG is transferred through the placenta, prob-

ably because of the presence of trophoblastic receptors that bind only IgG. Thus, although this physiologic process provides the newborn with a passive defense mechanism against different pathogens, it can also have some disadvantages in certain pathologic conditions. Because trophoblasts bind and engulf IgG molecules, transport them across the cell, and release the antibody on the fetal side, passage of maternal IgG to the fetus in cases of maternal Graves' disease, myasthenia gravis, and autoimmune thrombocytopenic purpura might cause transient manifestations of the disease in the newborn. Approximately 65% of our patients were studied because of suspected fetal hemolytic disease. Although only IgG1 and IgG3 are thought to be associated with this disorder (because only IgG1 and IgG3 have an Fc portion that can effectively bind with the phagocytic IgG Fc receptor) (1), we could not find any differences between the IgG subclass concentrations in the fetuses with suspected hemolytic disease and those in the other fetuses. This may indicate that despite the important clinical consequences of the passage of these antibodies, the amount of IgG1 and IgG3 directed against erythrocytes is quantitatively insignificant in patients with hemolytic disease of the newborn.

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