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THE TRANSFER OF HUMAN IgG SUBCLASSES FROM MOTHER TO FOETUS

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SUMMARY

The concentration of IgG subclasses was measured in matched pairs of maternal and cord serum by single radial immunodiffusion. IgG1, IgG3 and IgG4 were present in similar amounts in both the mothers and infants. IgG2 was present in the mothers at about 3 times the amount in the cord blood.

INTRODUCTION

The human foetus has a level of IgG very similar or marginally higher than that of the mother. Studies on babies born to hyper- or hypo-gammaglobulinaemic mothers have shown that the level of immunoglobulin found in the infant depends closely on that in the mother (Senecal & Berton, 1957; Zak & Good, 1959; Bridges *et al.*, 1959). Furthermore, most of the infant's immunoglobulin carried allotypic determinants derived from the mother rather than from the child (Grubb & Laurell, 1956; Linnet-Jepsen, Galatius-Jepsen & Hauge, 1958). However, Gm typing shows that the foetus produces some endogenous IgG but only in very small quantities (Martensson & Fudenberg, 1965). IgM, IgA and IgD (Rowe & Fahey, 1965) do not appear to cross the placenta.

Within the IgG class some selective transfer of different Gm groups has been found in some individuals, but this has not been determined quantitatively (Hunger & Thierbach, 1963; Ropartz, Rivat & Rousseau, 1965). Because of these findings and the known differences between IgG subclasses in their biological properties it was decided to investigate the presence of these subclasses quantitatively in matched pairs of maternal and foetal sera.

MATERIALS AND METHODS

Serum samples. Venous blood was obtained from mothers at the same time as cord blood from infants. Serum was collected after clotting, and this was stored at $-20°C$ until testing within 2 weeks.

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TABLE 1. IgG Subclasses in maternal and cord sera

	No. tested	IgG1	IgG2	IgG3	IgG4
Mother	20	73±22	144±99	117±38	82±37
Foetus	20	80±24	60±27	125±37	89±51

Results given as % mean (± standard deviation) relative to the standard pool of serum.

Antisera. Antisera to human IgG subclasses were prepared by immunizing rabbits with whole IgG myeloma protein. The myeloma proteins were separated from serum using Pevikon block electrophoresis. The separated proteins were then further purified by isoelectric focussing using the LKB sucrose gradient system.

The purified myeloma proteins were injected subcutaneously and intramuscularly in Freund's complete adjuvant, followed by repeated doses in Freund's incomplete adjuvant over a period of several months. All the antisera were made specific by absorption with K and L Bence Jones proteins, and with myeloma proteins of all the other subclasses. The antisera were checked for specificity by Ouchterlony immunodiffusion and compared with standard reagents from other laboratories.

Quantitation of immunoglobulin. The concentration of the subclasses present in the sera was determined by the single radial diffusion method of Mancini, Carbonera & Heremans (1964). A pool of serum from eight normal adults was used to provide the standard curve.

With the anti-IgG2 and anti-IgG1 no precipitation was seen after incubation. These plates were washed with cold phosphate buffered saline and then goat antirabbit serum was added. This produced rings of precipitation which could be measured.

RESULTS

The results obtained are shown in Table 1. These are expressed as a percentage of the concentration of the subclasses present in a pool of normal adult serum. There was only a small

TABLE 2. The relationship between maternal and foetal levels of IgG subclasses

	IgG1	IgG2	IgG3	IgG3
Maternal:* foetal ratio	0·93±0·22	2·58±1·93	0·94±0·19	0·96±0·25
P less than†	N.S.	0·001	N.S.	N.S.
Correlation coefficient	0·66	0·59	0·80	0·80
P less than	0·005	0·01	0·001	0·001

* The mean (± standard deviation) of the individual maternal:foetal ratios are given.

† Paired *t*-test.

variation between individuals in subclasses IgG1, 3 and 4. This was true also of IgG2 in the foetal sera but in the mothers there was a wide scatter of values, the coefficient of variance being 0·69 as compared with 0·30–0·45 for the other subclasses.

The maternal: foetal ratios are shown in Table 2. In IgG1, 3 and 4 there is a slightly higher concentration of immunoglobulin in the foetal sera than in the maternal. But this difference is not significant by the paired *t*-test. However, in IgG2 the position is reversed. Here there is a higher concentration of immunoglobulin in the maternal serum, almost 3 times as much, and this difference is significant ($2P<0·001$).

DISCUSSION

These results indicate that there is some selection in the transfer of IgG across the placenta in that the levels of IgG2 in the foetus are relatively low.

There is some evidence that the placenta is capable of acting as a selective filter for different IgG molecules. Ropartz *et al.* (1965) using Gm and InV typing found that in twenty-one out of eighty-one mother–offspring pairs, the cord blood failed to possess a factor present in the mother. The factors which mainly failed to cross were InV(2) and Gm(8). Gm(8) is an allotypic marker found on the gamma-1 heavy chain. However, this is not easy to reconcile with the finding that the other IgG1 allotypes tested were found to cross normally and that in our studies total IgG1 was virtually the same in maternal and foetal serum. Other workers have looked at the transfer of IgG1 using Gm(a) as a marker. Linnet-Jepson (1958) concluded on the basis of 165 pairs that 'the child is invariably born with the Gm group of his mother'. But more recently Hunger & Thierbach (1963) have shown on the basis of 500 pairs that only thirteen of these did not reflect the maternal Gm(a) pattern. However, these investigations were not carried out quantitatively so that how much crossed cannot be determined. The only other subclass tested for, by Ropartz, was IgG3 with Gm(5) allotype. This was found to pass to the foetus in most cases. Allotypes associated with IgG2 and IgG4 were not tested.

Very recently Wang *et al.* (1970) have published work in which they compared a *pool* of foetal sera with a pool of normal adult sera. They found, in agreement with us, that IgG3 and IgG4 were present in foetal serum but that the concentration of IgG2 was considerably lower than normal, as determined by Ouchterlony dilution.

That it is IgG2 which behaves differently is perhaps strange as on the amino acid sequence data it falls structurally between the IgG1-3 pair and IgG4. But as it is the Fc piece which is responsible for placental transfer (Gitlin *et al.*, 1964), this peptide could be examined further to see what sequence possibly provides the site for diaplacentary movement. Also this same structure may be responsible for transfer across other biological membranes.

It is possible that none of the IgG2 crosses the placenta and that the small amount present in the cord serum has been produced by the foetus. This might account for the weaker correlation between maternal and foetal levels of IgG2 than of the other subclasses. Of course, IgG2 may cross the placenta but then be removed by some mechanism such as increased catabolism in the foetus or selective binding to the placenta. To investigate this further it would be of value to examine the Gm grouping of the IgG2 to ascertain whether the small amount present in the cord blood is derived from the mother.

The reasons for the wide variation in IgG2 concentrations in the maternal sera remain unknown.

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