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# Postnatal decline of maternally acquired rubella antibodies

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## SUMMARY

The postnatal decline of maternally acquired rubella antibody was studied in a large group of infants. A high degree of variability was found in the rate of antibody decline (half-life). Ninety-two babies had rubella antibody half-lives lying between 14 and 70 days and three had values considerably higher. There was no significant difference between the rubella antibody half-lives of the sexes. The antibody titre at birth was weakly correlated with both birth weight and gestational age. There was a highly significant positive correlation between the baby's antibody titre at birth and that of its mother. There was a positive relationship between the half-life and the persistence of rubella antibody. Some babies had no detectable antibody by 2 months whereas others still possessed antibody at 9 months. It was found that the relationship between the half-life and the rubella antibody titre at or near birth could be described by a rectangular hyperbola.

## INTRODUCTION

A number of studies have shown that maternally acquired antibody in the newborn child decreases at an exponential rate after birth (Allansmith, McClellan, Butterworth & Maloney, 1968; Schultze & Heremans, 1966). Most of these studies have been related to total IgG immunoglobulin and few have been concerned with the rate of decline of specific antibodies.

An opportunity to study the decline of specific maternal antibody in a large group of newborn babies was recently given to us by an intensive longitudinal perinatal survey. A study was made of the decline of rubella antibody, in the first instance, for a number of reasons. First, a previous (unpublished) study by us had indicated that 79% of infants could be expected to have rubella antibody of maternal origin in their sera at birth. Secondly, a sensitive quantitative test (haemagglutination-inhibition test) had recently been developed for rubella antibody and this test was readily adaptable to the micro-system needed for the small volumes of sera available. A third and practical reason for studying the decline of rubella antibody was related to the diagnosis of congenital rubella infections. A positive serological diagnosis of congenital rubella is indicated by the failure of the affected infant's antibody titre to decline in a 'normal' manner following birth. However, to our knowledge, no detailed studies of the 'normal' decline of rubella antibody had been reported.

This paper reports not only the decline of maternally acquired rubella antibodies in a large group of babies, but also the relationship between the rates of antibody decline and persistence of antibody and initial antibody titre.

## MATERIALS AND METHODS

### *Study group*

The babies in this study were born between April 1967 and June 1968 at the Royal Hospital for Women, Paddington, N.S.W. Of the 120 babies in the original group, 95 (79%) possessed rubella antibody; these composed the study group. The birth weights and gestational ages of the babies are summarized in Tables 1 and 2.

Table 1. *Classification of the study group by birth weight*

| Birth weight (g.) | No. of babies in group | Birth weight (g.) | No. of babies in group |
|---|---|---|---|
| 1100–1500 | 12 | 3001–3500 | 5 |
| 1501–2000 | 28 | 3501–4000 | 5 |
| 2001–2500 | 27 | Total | 95 |
| 2501–3000 | 18 | | |

Table 2. *Classification of the study group by gestational age*

| Gestational age (weeks) | No. of babies in group | Gestational age (weeks) | No. of babies in group |
|---|---|---|---|
| 28–30 | 6 | 39–42 | 31 |
| 31–34 | 23 | 43–44 | 5 |
| 35–38 | 30 | Total | 95 |

### *Sera for the study*

Blood was taken from the mothers at parturition, from the babies at birth or within 10 days following birth ('initial serum') and also at regular intervals for a period of 2–12 months. Generally four to six sera were collected from each infant. Serum was removed from the clotted blood and was stored at $-30°$ C.

### *Serological techniques*

Rubella antibody was determined by the haemagglutination-inhibition (HI) test (Stewart *et al.* 1967), modified in that non-specific inhibitors were removed from the sera by manganese chloride and heparin (Cooper, Matters, Rosenblum & Krugman, 1969), that the sera were treated with pigeon red blood cells to remove naturally occurring agglutinins and that 0·2% pigeon cells were used in the test.

In order to minimize bias, the babies whose sera were to be tested on a given day were selected by an appropriate ranking procedure and all the sera from a given baby were tested at the same time. Ten samples of a standard positive serum were included in each test as a check on the within-test and between-test variability.

Each baby in the study group was found to possess detectable antibody in at least three but usually four to six sequential sera. The antibody titres were punched onto data cards and were analysed in an IBM 360 Model 50 computer.

Using these antibody titres, a linear regression of log. titre against time (days from birth) was computed, and the print-out included the rate of rubella antibody decline (half-life) for each baby.

## RESULTS

### *Rate of antibody decline (half-life)*

A frequency distribution of the half-life values obtained is shown in Fig. 1. Half-lives ranged from 14 to 259 days with a mean of 43 days and a mode of 36 days. In most cases the straight line obtained from the regression equation was a good fit to the observed values. With six babies, including the baby with a rubella antibody half-life of 259 days, a twofold or greater rise or fall in rubella antibody titre, as compared to the titre at birth, took place during the first month of life prior to the commencement of exponential antibody decline. In addition, four babies had a stationary antibody titre for between 1 and 2 months of life, after which antibody declined in an exponential manner.

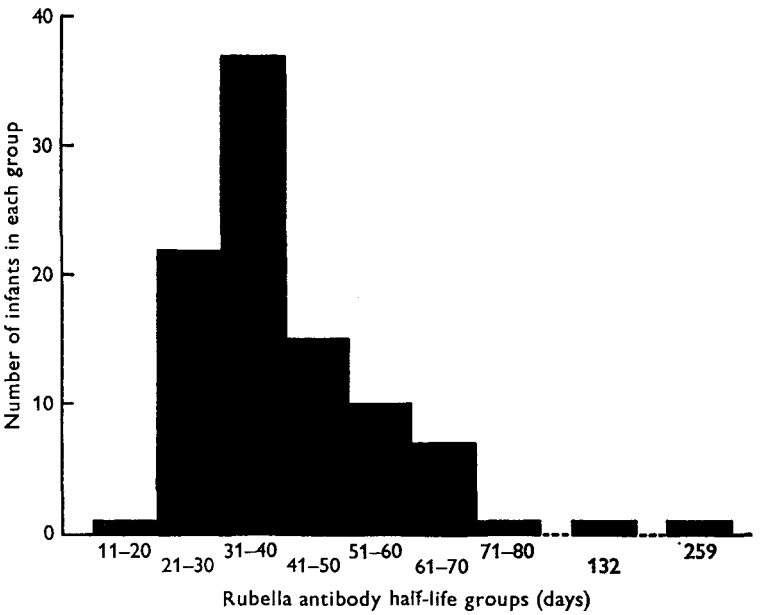


Fig. 1. Frequency distribution of rubella half-life values.

### *Antibody status in twins*

There were 10 pairs of twins in the original group. The antibody status (i.e. presence or absence of antibody) of both twins in a pair was always identical. Seven pairs were antibody positive. In these pairs, not only was the titre of rubella antibody at birth similar for both twins of a pair, but the half-lives of rubella antibody in both twins were also similar.

*Sex of the babies*

There was no statistically significant difference between the mean rates of antibody decline of the sexes. The frequency distributions of half-lives in the two sexes were almost identical.

*Relationships between initial antibody titre, birth weight and gestational age*

There was a positive correlation between initial antibody titre and birth weight and between initial antibody titre and gestational age. In both cases there was a considerable scatter of points and the relationships were only weakly significant ($P = 0{\cdot}05$).

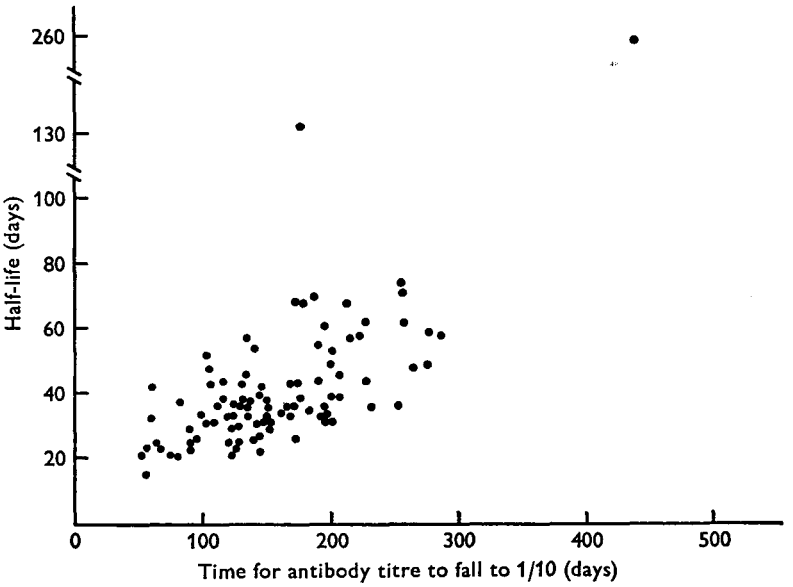


Fig. 2. Relationship between half-life and time calculated for rubella antibody to decline to a titre of 1/10.

*Relationship between antibody titres of infants at birth and those of their mothers*

There was a highly significant linear correlation between the baby's initial rubella antibody titre and that of its mother ($r = 0{\cdot}99$, $P < 0{\cdot}001$). Only 8% of the babies had a titre twofold greater or lower than that of their mothers, and these differences were never greater than sixfold.

*Duration of detectable maternally acquired rubella antibody*

The lowest titre of rubella antibody detectable by our test system was 1/10. In order to obtain some idea of the persistence of rubella antibody, the number of days from birth for the titre to fall to 1/10 was calculated from the regression equation for each baby. The relationship between half-life and the number of days for the titre to reach a level of 1/10 for each baby is shown in Fig. 2. It can be seen that there is a positive relationship between half-life and the duration of antibody. In addition, it was observed that 17 babies (18%) had detectable antibody at 6 months of age and seven (5%) had antibody at 9 months.

*Relationship between half-life and initial antibody titre*

The decline of maternally acquired antibodies in the infant is a reflexion of the catabolism of IgG. It has been shown that IgG catabolism is related to the IgG concentration in the serum such that increasing concentrations of IgG lead to an increase in its catabolism and hence a decrease in its half-life (Brambell, Hemmings & Morris, 1964; Brambell, 1966). It has been postulated that this relationship between half-life and concentration of IgG takes the form of a rectangular hyperbola.

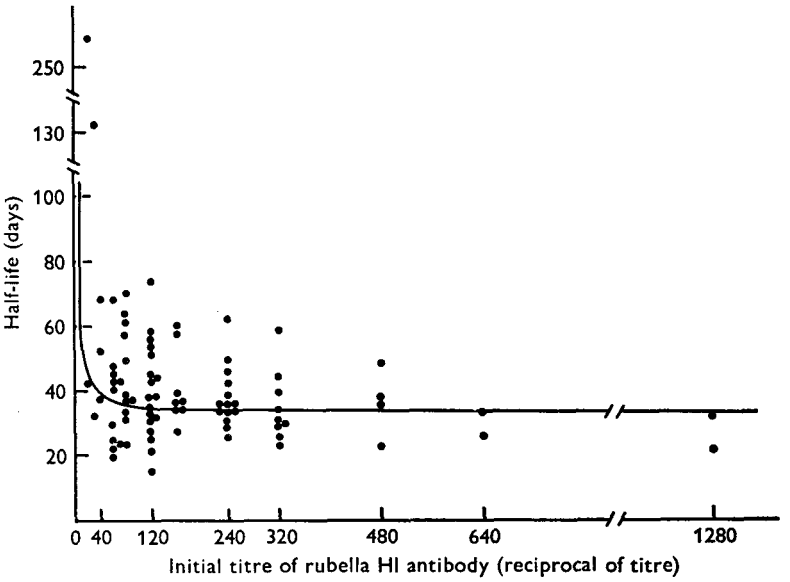


Fig. 3. Relationship between half-life and initial titre of rubella antibody in infants. The curve (Brambell *et al.* 1964) is

$$\text{half-life} = \frac{\text{initial titre ln } 2}{0{\cdot}02\ (\text{initial titre} - 3{\cdot}25)}$$

In this study the relationship between the rate of decline of an IgG antibody of a given specificity and its titre in the infants' serum at birth was determined. The relationship between the half-life of rubella antibody in these babies and the titre of antibody at birth could be described by a rectangular hyperbola (Fig. 3). Few of the babies had an initial titre of 1/40 or less and consequently few points were obtained at the lower end of the antibody axis. The variability between babies over the range of initial antibody titres was not constant and increased at the lower end of the antibody axis. For this reason, the linear equivalent of the model proposed by Brambell *et al.* (1964) (see Fig. 3) was used in this study. It was found that there was a statistically significant relationship ($P = 0{\cdot}01$) of the type described. Although the measurement errors of initial titre and half-life are correlated, it was found that this correlation had a negligible effect on the significance of the relationship between half-life and initial titre.

## DISCUSSION

The relationship noted here between the antibody titres in the mother and infant at birth has been previously reported both for total IgG and also for other specific antibodies (Schultze & Heremans, 1966). Similarly, a relationship between IgG levels and both birth weight and gestational age has been demonstrated (Hobbs & Davis, 1967; Jones, 1969). This study simply serves to extend these relationships to rubella antibody in a large series of children.

On the other hand, there are two aspects of this study which are of considerable importance. First, it is noteworthy that there has been a high degree of variability between babies in the rate of decline of maternally acquired rubella antibody. To our knowledge, this has not been previously reported. The half-life of maternally acquired antibody in infants has usually been considered as lying between 20 and 40 days. This has applied to total IgG as well as to a wide range of viral antibodies (Schultze & Heremans, 1966) including polio antibody (Perkins, Yetts & Gaisford, 1958), measles antibody (Strauss & Zeman, 1967), and rubella antibody (Vesikari, Vaheri, Pettay & Kunnas, 1969).

The observations recorded above with this large group of babies indicate that the upper limit of the half-life range must now be extended to at least 70 days and possibly even higher (Fig. 1). Within this group of children there were babies who had lost all detectable antibody by 2 months of age and others who still possessed detectable antibody at 9 months. As rubella antibody undoubtedly declines to titres below the level of detectability of the HI test, it is probable that in many babies such antibody persists beyond 9 months. It has been suggested that the time for the complete disappearance of herpes simplex antibody of maternal origin was from 3 to 15 months (Anderson & Hamilton, 1949), and for group-B arbovirus antibody, 6 months or longer (Carey, Myers, Wilson & Manoharan, 1968).

The wide range of rubella half-lives obtained in this study indicates that caution must be exercised in the interpretation of antibody titres in the serological diagnosis of congenital rubella infections. It is doubtful whether laboratory confirmation of the diagnosis of congenital rubella can be made solely on the presence of rubella antibody at 6 or 9 months of age. This criterion has occasionally been used in the confirmation of a diagnosis (Hardy, McCracken, Gilkeson & Sever, 1969; McCracken *et al.* 1969).

It was not considered likely that babies in this series with long half-lives had a congenital rubella infection, since none of the babies had any of the known congenital abnormalities of *in utero* rubella infection. In addition, in all 12 babies having a half-life of 60 days or greater, the antibody level eventually reached a titre of less than 1/10. In congenital rubella the antibody titre does not decline during the first 6 to 12 months of life (Michaels, 1969; Vesikari *et al.* 1969), although it may start to decline at 15 to 18 months of age (Hardy *et al.* 1969). Similarly, it was not considered that the long half-lives encountered were due to the presence of non-specific inhibitors in some of the sera. All the results obtained, including those from the negative control sera, indicated that all of the inhibitors had been removed from the sera. Little is known about the host factors which affect the

rate of decline of passively acquired antibodies, although it has been suggested that the basal metabolic rate may influence their catabolism (Solomon, Waldmann & Fahey, 1963; Schultze & Heremans, 1966).

The second finding of considerable importance is the relationship between half-life and initial titre. There have been a number of studies which have suggested that the catabolism of passively acquired IgG is related to its concentration in the serum in such a way that as the IgG concentration is increased the rate of catabolism also increases (Brambell *et al.* 1964; Brambell, 1966). Such a phenomenon has been reported for a number of species including the mouse (Fahey & Robinson, 1963; Fahey & Sell, 1965), the rabbit (Andersen & Bjørneboe, 1964) and man (Solomon *et al.* 1963; Waldmann & Schwab, 1965; Waldmann, 1969). Although most of these studies have been carried out in adults, it has been found that the half-life of passively administered gammaglobulin in children with agammaglobulinaemia was 50–60 days whereas, in normal children, it was 20–40 days (Gitlin, Janeway, Apt & Craig, 1959).

Although this study has been with antibody of one specificity, as distinct from total IgG, these results are in reasonable agreement with the model postulated by Brambell *et al.* (1964). To our knowledge, these results represent the first detailed study in infants of the relationship between the postnatal decline of maternally acquired antibody of a given specificity and the titre of such antibodies at birth. The results suggest that with a specific antiviral antibody, namely rubella antibody, babies having high titres at birth lose their antibody rapidly, whereas in babies with low initial titres, antibody declines over a longer period of time.

Precise information such as this on the postnatal decline of specific maternally acquired antibodies is important for several reasons. Maternal antibody in the infant may provide protection against infection (Anderson & Hamilton, 1949), it may affect the efficacy of vaccination in infants (Perkins *et al.* 1958) and it has recently been suggested by Chanock *et al.* (1968) that it may even be a factor in the pathogenesis of certain virus infections. Future studies will be carried out using different antigens with the same sera in an attempt to determine whether other antiviral antibodies, especially viral respiratory ones, decline in the same manner as rubella antibody. An attempt will also be made to determine whether the half-life of antibody of a given specificity depends entirely on its own concentration in the serum or whether it is influenced by the level of other serum antibodies.

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## REFERENCES


Allansmith, M., McClellan, B. H., Butterworth, M. & Maloney, J. R. (1968). The development of immunoglobulin levels in man. *Journal of Pediatrics* **72**, 276.

Andersen, S. B. & Bjørneboe, M. (1964). Gammaglobulin turnover in rabbits before and during hyperimmunization. *Journal of Experimental Medicine* **119**, 537.

Anderson, S. G. & Hamilton, J. (1949). The epidemiology of primary herpes simplex infection. *Medical Journal of Australia* **1**, 308.

Brambell, F. W. R., Hemmings, W. A. & Morris, I. G. (1964). A theoretical model of $\gamma$-globulin catabolism. *Nature, London* **203**, 1352.

Brambell, F. W. R. (1966). The transmission of immunity from mother to young and the catabolism of immunoglobulins. *Lancet* ii, 1087.

Carey, D. E., Myers, R. M., Wilson, E. & Manoharan, A. (1968). Transplacentally-acquired group-B HI antibody and dengue infection among infants in Vellore, South India. *Indian Journal of Medical Research* **56**, 1468.

Chanock, R. M., Parrott, R. H., Kapikian, A. Z., Kim, H. W. & Brandt, C. D. (1968). Possible role of immunological factors in pathogenesis of RS virus lower respiratory tract disease. *Perspectives in Virology* **6**, 125.

Cooper, L. Z., Matters, B., Rosenblum, J. K. & Krugman, S. (1969). Experience with a modified rubella haemagglutination-inhibition antibody test. *Journal of the American Medical Association* **207**, 89.

Fahey, J. L. & Robinson, A. G. (1963). Factors controlling serum $\gamma$-globulin concentration. *Journal of Experimental Medicine* **118**, 845.

Fahey, J. L. & Sell, S. (1965). The immunoglobulins of mice. V. The metabolic (catabolic) properties of five immunoglobulin classes. *Journal of Experimental Medicine* **122**, 41.

Gitlin, D., Janeway, C. A., Apt, L. & Craig, J. M. (1959). *Cellular and Humoral Aspects of the Hypersensitive State*, p. 375. Ed. H. Sherwood Lawrence. New York: Hoeber-Harper.

Hardy, J. B., McCracken, G. H., Gilkeson, M. R. & Sever, J. L. (1969). Adverse fetal outcome following maternal rubella after the first trimester of pregnancy. *Journal of the American Medical Association* **207**, 2414.

Hobbs, J. R. & Davis, J. A. (1967). Serum $\gamma$G-globulin levels and gestational age in premature babies. *Lancet* i, 757.

Jones, W. R. (1969). Immunoglobulins in fetal serum. *Journal of Obstetrics and Gynaecology of the British Commonwealth* **76**, 41.

Michaels, R. H. (1969). Immunologic aspects of congenital rubella. *Pediatrics* **43**, 339.

McCracken, G. H., Hardy, J. B., Chen, T. C., Hoffman, L. S., Gilkeson, M. R. & Sever, J. L. (1969). Serum immunoglobulin levels in newborn infants. II. Survey of cord and follow-up sera from 123 infants with congenital rubella. *Journal of Pediatrics* **74**, 383.

Perkins, F. T., Yetts, R. & Gaisford, W. (1958). Serological response of infants to poliomyelitis vaccine. *British Medical Journal* ii, 68.

Schultze, H. E. & Heremans, J. F. (1966). *Molecular Biology of Human Proteins*, vol. 1. *Nature and Metabolism of Extracellular Proteins*. Amsterdam: Elsevier.

Solomon, A., Waldmann, T. A. & Fahey, J. L. (1963). Metabolism of normal 6·6S $\gamma$-globulin in normal subjects and in patients with macroglobulinemia and multiple myeloma. *Journal of Laboratory and Clinical Medicine* **62**, 1.

Stewart, G. L., Parkman, P. D., Hopps, H. E., Douglas, R. D., Hamilton, J. P. & Meyer, H. M. (1967). Rubella-virus haemagglutination-inhibition antibody test. *New England Journal of Medicine* **276**, 554.

Strauss, J. & Zeman, L. (1967). Study of measles neutralizing antibody levels in children in first months of their life. *Journal of Hygiene, Epidemiology, Microbiology and Immunology* **11**, 40.

Vesikari, T., Vaheri, A., Pettay, O. & Kunnas, M. (1969). Congenital rubella: Immune response of the neonate and diagnosis by demonstration of specific IgM antibodies. *Journal of Pediatrics* **75**, 658.

Waldmann, T. A. & Schwab, P. J. (1965). IgG (7S gamma globulin) metabolism in hypogammaglobulinemia: Studies in patients with defective gamma globulin synthesis. *Journal of Clinical Investigation* **44**, 1523.

Waldmann, T. A. (1969). Disorders of immunoglobulin metabolism. *New England Journal of Medicine* **281**, 1170.