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Half-Lives of Immunoglobulins IgG, IgA and IgM in the Serum of New-Born Pigs

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Summary. Half-lives of immunoglobulins IgA, IgG and IgM labelled with Iodine-125 were measured and compared with the half-lives of these immunoglobulins calculated from plasma levels during the first 2–3 weeks of life. The two methods gave similar results for IgG and IgA, but the mean IgM half-life calculated from plasma IgM levels was significantly longer than the mean half-life of IgM-I^{125}. IgM production and passage into the circulatory system during the first week of life could account for this difference.

INTRODUCTION

The young pig is born virtually devoid of immunoglobulins since, under normal circumstances, they do not cross the placental barrier. Low levels of a protein related to IgG are detectable in piglet serum at birth (Franek and Riha, 1964; Prokesova, Rejnek, Sterzl and Travnicek, 1969; Porter, 1969; Bourne, 1971) and trace amounts of IgA and IgM occur in some presuckling serum samples (Curtis and Bourne, 1971). In the first 24–36 hours of life, colostral immunoglobulins are absorbed through the small intestine of the piglet as intact molecules and give rise to high circulatory immunoglobulin levels which reach a peak 24 hours after birth (Porter and Hill, 1970; Curtis and Bourne, 1971). Serum IgG and IgA levels decrease sharply in the second day of life and this could be due to the equilibration of absorbed IgG and IgA between intra- and extra-vascular fluids (Schultz and Heremans, 1966). After 2 days of age, serum immunoglobulin levels fall exponentially.

Immunoglobulin half-lives in the serum of pigs were calculated from serum immunoglobulin levels during the first weeks of life (Porter and Hill, 1970; Curtis and Bourne, 1971). Immunoglobulin production by the young pig will contribute to circulatory levels and so apparent half-lives calculated from serum immunoglobulin levels may be longer than the immunoglobulin half-lives actually are.

In this study, the half-lives of administered ^{125}iodine-labelled immunoglobulins were measured and compared with half-lives calculated from serum immunoglobulin levels.

MATERIALS AND METHODS

Preparation of immunoglobulins

IgA was prepared from colostrum by the method described by Bourne (1969). Serum IgM was prepared by the method of Bourne (1971).

Colostral IgG

Colostral whey was fractionated by reverse-flow exclusion chromatography on Sephadex G-200 (column size 10 × 100 cm) after extensive dialysis against 0·1 M Tris-HCl 1·0 M NaCl buffer (pH 7·4) which was used for elution. Five peaks were found by monitoring at 280 nm (Fig. 1). Fractions from the top of the third (7S) peak were used for the preparation of colostral IgG by the method described for serum IgG complex (Curtis and Bourne, 1971).

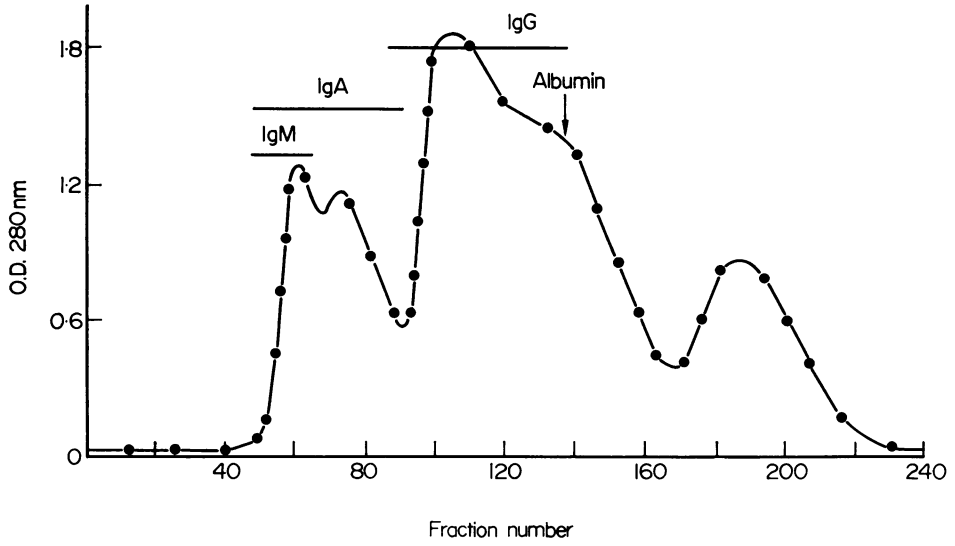

FIG. 1. Gel-filtration run on Sephadex G-200 of sow's colostrum.

Trace-labelling

Immunoglobulins were trace-labelled with ^{125}iodine-(approximately 20 μCi/mg) by the Chloramine-T method of Hunter and Greenwood (1962) as described by Freeman (1966). Labelled immunoglobulin was mixed with unlabelled immunoglobulin to give a final specific activity of approximately 5 μCi/mg.

Administration of labelled immunoglobulin

In the first litter studied, IgG-^{125}I was administered to new-born pigs by two routes; either 10–20 mg were given intravenously, or 20–30 mg by mouth. The route of administration did not affect the half-life of IgG and so a second litter was dosed by mouth during the period of maximum immunoglobulin absorption, i.e. within 12 hours of birth. IgA-^{125}I was administered by the oral route only. One litter was given IgM-^{125}I by mouth, but only low levels of activity were found in the piglets' sera and so subsequent litters were injected intravenously with 5–10 mg of IgM-^{125}I. For IgA and IgG half-life studies, the first blood samples were collected 2 days after dosing to allow the equilibration of administered immunoglobulin to occur. For IgM studies, the first blood sample was taken 24 hours after dosing. Subsequent samples were collected at 2-day intervals for 10 days (for IgA and IgM) and to 5 weeks (for IgG).

Treatment of blood samples

Blood samples (2·0–2·5 ml) were collected into heparinized tubes and the plasma was

separated immediately by centrifugation. Proteins were precipitated from 1 ml of plasma with 10 per cent trichloroacetic acid and the radioactivity of the precipitates was counted in a γ-ray counter (Panax Nucleonic Instruments).

Immunoglobulin levels in the plasma samples were measured by the radial immunodiffusion technique of Fahey and McKelvey (1965).

Calculations

Immunoglobulin concentrations in the serum of young pigs fall exponentially until the disappearance of absorbed colostral immunoglobulins is balanced by the production, by the young pig, of its own circulatory immunoglobulins. The half-lives of absorbed colostral immunoglobulins were measured over the period of exponential decrease in concentration, assuming that the equilibration of immunoglobulin was complete by 2 days of age.

The half-lives of ^{125}I-labelled immunoglobulins were calculated for each pig from the regression of log counts on time. The half-lives of absorbed colostral immunoglobulins in the same pigs were calculated from the regression of log plasma immunoglobulin concentration on time. Non-significant regressions were discarded.

Immunoglobulin levels in serum samples collected during the first 6 weeks of life from pigs in seven litters had previously been measured (Curtis and Bourne, 1971) and a mean immunoglobulin half-life for each litter was calculated from the regression of log immunoglobulin concentration (using data from every pig in the litter) on time.

RESULTS

Half-life of IgA

Half-lives of IgA in the plasma of individual pigs in three litters, calculated from the fall in radioactive counts in plasma-protein precipitates and from the fall in plasma IgA levels are given in Table 1. There was no significant difference between mean IgA half-lives measured by the two methods. There was a significant difference ($P<0{\cdot}05$) between litters in mean IgA half-life, calculated from the fall in levels of radioactivity, and between the mean IgA half-lives of litters V and W, calculated from plasma IgA levels.

Mean IgA half-lives of each of seven litters, calculated from serum IgA levels, are given in Table 2. The regression coefficients of log IgA concentration on time (from which the half-lives were calculated) were compared, and significant differences between regression coefficients and hence half-lives, are shown in Table 2.

Half-life of IgG

Half-lives of IgG in the plasma of individual pigs in two litters are given in Table 3. There was no significant difference between the mean IgG half-life calculated from the fall in radioactive counts in plasma protein precipitates and the mean IgG half-life calculated from plasma IgG levels. The former was calculated from data collected over 5 weeks, while the latter was calculated from IgG levels during the period of exponential decrease (2–19 days). There was a significant difference ($P<0{\cdot}01$) between the mean IgG half-lives of the two litters calculated from the fall in radioactivity in plasma protein precipitates.

Mean litter IgG half-lives in seven litters calculated from serum IgG levels are given in Table 4. The regression coefficients of log IgG concentration on time were compared and significant differences between regression coefficients are indicated in Table 4.

TABLE 1

IgA HALF-LIVES IN INDIVIDUAL PIGS CALCULATED FROM THE FALL IN IgA-^{125}I IN PLASMA PROTEIN PRECIPITATES AND PLASMA IgA CONCENTRATIONS (mg/100 ml)

Pig. No.	IgA half-life (days)	
	IgA-^{125}I	Plasma IgA Concentration
U 11	3·3	
U 12	3·2	
U 13	3·6	
U 14	2·7	
Litter mean	3·2	2·1*
V 2	2·7	3·0
V 3	2·8	2·7
V 4	2·7	2·8
V 5	2·8	3·3
V 6	2·6	2·9
V 11	2·5	2·7
V 12		2·9
V 13		2·6
Litter mean	2·7	2·9
W 3	2·3	2·0
W 4	2·5	2·1
W 5	2·2	2·1
W 6	2·3	2·1
W 14		2·1
W 15		1·9
Litter mean	2·3	2·1
Mean	2·7	2·5

* Mean IgA half-life calculated from plasma IgA levels of all four pigs.

TABLE 2

MEAN LITTER HALF-LIVES OF IgA IN SEVEN LITTERS, CALCULATED FROM SERUM IgA CONCENTRATIONS (mg/100 ml)

Litter No.	Regression coefficient of log IgA concentration (mg/100 ml) on time	Mean litter IgA half-life (days)
1	$-0{\cdot}142^{a}$	2·1
3	$-0{\cdot}132^{ab}$	2·3
4	$-0{\cdot}123^{bc}$	2·4
2	$-0{\cdot}112^{cd}$	2·7
6	$-0{\cdot}106^{cd}$	2·8
5	$-0{\cdot}104^{d}$	2·9
7	$-0{\cdot}101^{d}$	3·0
Mean		2·6

^{abcd} Figures with the same superscript are not significantly different and figures with different superscripts are significantly different ($P < 0{\cdot}05$).

TABLE 3

IgG HALF-LIVES IN INDIVIDUAL PIGS, CALCULATED FROM THE FALL IN IgG-^{125}I IN PLASMA PROTEIN PRECIPITATES AND PLASMA IgG CONCENTRATIONS (mg/ml)

Pig No.	IgG half-life (days)	
	IgG-I^{125}	Plasma IgG concentration
I 2*	7·6	
I 4†	6·8	
I 8†	8·4	
I 10*	6·5	
Litter mean	7·3	
T 11	11·2	11·4
T 12	12·1	12·4
T 13	9·3	9·6
T 14	8·7	8·5
T 15	10·6	9·8
T 16	10·3	9·3
T 17	8·8	9·7
Litter mean	10·1	10·1
Mean	9·1	10·1

* Dosed by mouth. † Injected intravenously.

Half-life of IgM

A litter of pigs which were given IgM-^{125}I by mouth had very low serum counts. There were no significant regressions of log counts on time and so these results are omitted. Pigs in three other litters were injected intravenously with IgM-^{125}I. In the third of these litters (litter P) it was obvious that the IgM-^{125}I in the serum of one pig (P 11) was not equilibrated when the first blood sample was taken, 24 hours after dosing (Fig. 2). IgM-^{125}I half-lives in all the pigs studied were recalculated omitting data from samples taken 24 hours after dosing. In all but one case the recalculated half-lives were longer, which suggests that the equilibration of IgM-^{125}I was not complete when the first blood sample was taken. The recalculated IgM-^{125}I half-lives are given in Table 5.

IgM half-lives calculated from plasma IgM concentrations are given in Table 5.

TABLE 4

MEAN LITTER HALF-LIVES OF IgG IN SEVEN LITTERS, CALCULATED FROM SERUM IgG CONCENTRATIONS (mg/ml)

Litter No.	Regression coefficient of log IgG concentration (mg/ml) on time	Mean litter IgG half-life (days)
4	$-0{\cdot}0456^{a}$	6·6
7	$-0{\cdot}0289^{ab}$	10·4
3	$-0{\cdot}0259^{b}$	11·6
1	$-0{\cdot}0224^{bc}$	13·4
2	$-0{\cdot}0206^{bc}$	14·6
5	$-0{\cdot}0165^{c}$	18·2
6	$-0{\cdot}0137^{bc}$	22·0
Mean		13·8

^{abc} Figures with the same superscript are not significantly different and figures with different superscripts are significantly different ($P < 0{\cdot}05$).

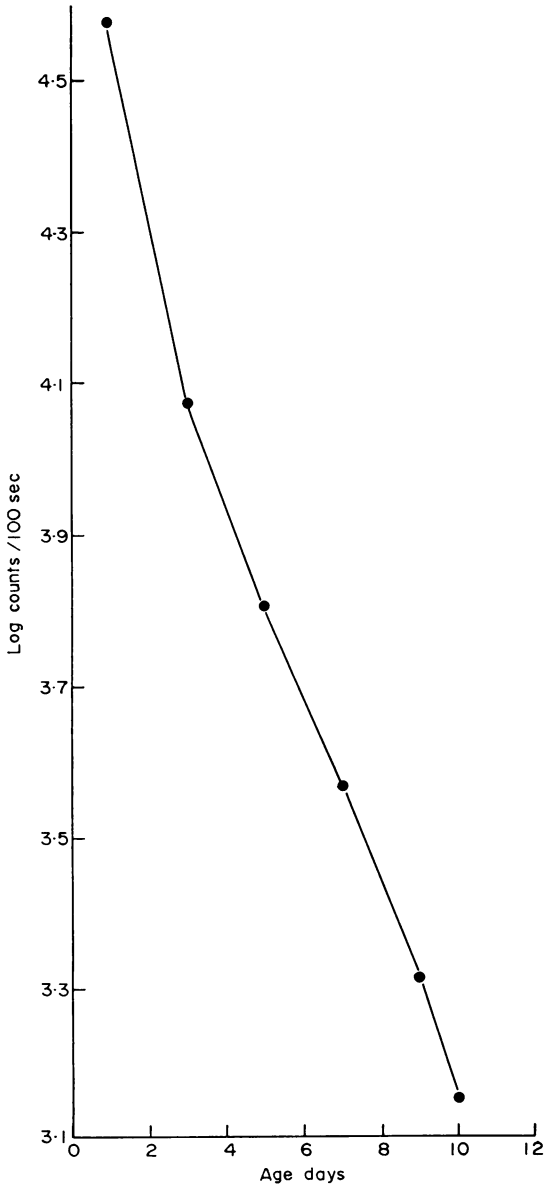

FIG. 2. The decrease with time of radioactivity (log counts/100 seconds) in plasma protein precipitates; pig P 11.

TABLE 5

IgM HALF-LIVES IN INDIVIDUAL PIGS, CALCULATED FROM THE FALL IN IgM-^{125}I IN PLASMA PROTEIN PRECIPITATES AND PLASMA IgM CONCENTRATIONS (mg/100 ml)

Pig No.	IgM half-life (days)	
	IgM-^{125}I	Plasma IgM concentration
A 1	3·1	4·1
A 2	2·8	—
Litter mean	3·0	—
M 1	3·1	5·0
M 2	2·6	4·3
M 3	2·8	6·6
M 9	2·7	4·0
M 10	3·0	N.S.
Litter mean	2·8	5·0
P 11	2·3	N.S.
P 12	2·5	N.S.
P 13	3·0	N.S.
P 14	2·8	N.S.
Litter mean	2·7	—
Mean	2·8	4·8

N.S. = Non-significant regression.

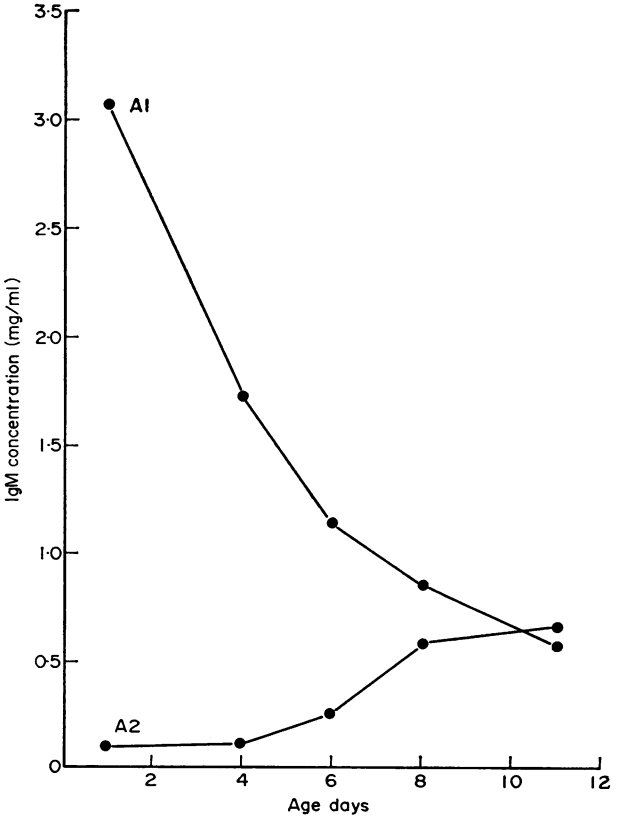

FIG. 3. IgM concentrations (mg/ml) in the plasma of pigs A 1 and A 2.

Plasma IgM levels of the pigs in litter P decreased from 1–5 days and started to rise again at 7 days of age. There were no significant regressions of log plasma IgM concentration on time in this litter. Pig A 2 did not appear to have taken very much colostrum since the IgM concentration in its plasma at 24 hours was very low compared with the IgM concentration in the plasma of the other pig studied in the same litter, A 1. The plasma IgM

TABLE 6

MEAN LITTER HALF-LIVES OF IgM IN FOUR LITTERS, CALCULATED FROM SERUM IgM CONCENTRATIONS (mg/100 ml)

Litter No.	Regression coefficient of log IgM concentration (mg/100 ml) on time	Mean litter half-life of IgM (days)
5	$-0{\cdot}0847^{a}$	3·6
6	$-0{\cdot}0643^{ab}$	4·7
7	$-0{\cdot}0558^{ab}$	5·4
1	$-0{\cdot}0474^{b}$	6·4
Mean		5·0

[ab] Figures with the same superscript are not significantly different and figures with different superscripts are significantly different ($P<0{\cdot}05$).

level of pig A 2 rose after 4 days of age and was similar to that of pig A 1 at 11 days (Fig. 3).

There was a highly significant difference ($P<0{\cdot}001$) between the mean IgM half-life calculated from plasma IgM levels and the mean half-life of IgM-^{125}I.

Mean litter IgM half-lives of four litters were calculated from serum IgM levels (Table 6). Significant differences between regression coefficients are indicated.

DISCUSSION

IgA immunoglobulin has a half-life in new-born pig serum of 2–3 days. The half-lives calculated from serum IgA levels were similar to the half-lives of administered IgA-^{125}I. This suggests that IgA production by pigs during the first 7–12 days of life does not contribute significantly to serum IgA levels. IgG half-lives in piglet serum range from 6·5 to 22·5 days. As with IgA, the half-lives of administered IgG-^{125}I were similar to those calculated from serum IgG levels, suggesting that any IgG production by the young pig in the first 2–3 weeks of life contributes little to serum IgG levels.

Significant differences in mean IgA and IgG half-lives between litters were found. The disappearance of immunoglobulin from the serum of the young pig is due to catabolism and dilution caused by increased body size and hence increased blood volume. Since catabolic rate is less likely than growth rate to vary significantly between litters, the more rapid disappearance of immunoglobulins from the serum of pigs in some litters is probably a reflection of the higher growth rate of these pigs. The ranking of litters 1–7 in Tables 2 and 4 in terms of half-lives of IgA and IgG, suggests that litters 1, 3 and 4 may have had a higher mean growth rate than litters 2, 5 and 6.

Administered IgM-^{125}I had a half-life of 2·5–3 days in the serum of new-born pigs. Apparent half-lives of IgM calculated from serum and plasma IgM levels ranged from 3·5 to 6·5 days. This suggests that IgM production must be contributing significantly to IgM circulatory levels in the first 10–12 days of life. Porter and Hill (1970) could not

detect IgM in the serum of two colostrum-deprived pigs until after the 20th day of life, but in a litter of suckled pigs serum IgM levels decreased to a minimum at 5 days, then started to rise again at 7 days of age. In the present study, IgM levels in the sera of the pigs in litter P started to rise again at 7 days of age and the IgM level in the serum of pig A 2, which appeared to have taken very little colostrum, started to rise after 4 days of age. The production of IgM and its passage into the circulatory system in significant amounts, therefore, does appear to occur in the first week of life in the suckled pig.

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