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The Transfer of Immunoglobins IgG, IgA and IgM from Serum to Colostrum and Milk in the Sow

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Summary. The proportion of IgA, IgG and IgM in sow colostrum and milk that is derived from serum has been determined by measuring the transfer of ^{125}I-labelled immunoglobulin from serum. All colostral IgG and a high proportion of IgM are derived from serum as is 40 per cent of IgA. Thus it would appear that colostrum is not a true secretion since 90 per cent of its immunoglobulin content is of serum origin.

In milk, however, 90 per cent of IgA and IgM and 70 per cent of IgG would appear to be locally produced in mammary tissue.

INTRODUCTION

Antibody production in mammary tissue, following local stimulation, has been demonstrated in the rabbit (Batty and Warrack, 1955), cow (Mitchell, Walker and Bannister, 1953), sheep (Lascelles, Outteridge and Mackenzie, 1966; Outteridge and Lascelles, 1967) and goat (Mitchell, Guerin and Pasieka, 1967). More recently, attempts have been made to identify the immunoglobulins involved. Genco and Taubman (1969) have shown that IgA immunoglobulin is formed locally in rabbit mammary tissue, following local antigenic stimulation. Eddie, Schulkind and Robbins (1971) suggested that all three classes of immunoglobulin are produced in rabbit mammary tissue. The formation of IgA following local stimulation has been demonstrated in sheep mammary tissue (Lascelles and McDowell, 1970) and fast IgG and IgM antibodies are also present in milk from stimulated glands (Outteridge, Mackenzie and Lascelles, 1968).

The main immunoglobulin of sows' milk is IgA (Porter, Noakes and Allen, 1970; Curtis and Bourne, 1971) which although present in colostrum is only a minor component, IgG constituting over 80 per cent of the total colostral immunoglobulin (Porter, 1969; Curtis and Bourne, 1971).

The work reported here is part of a study of local immunoglobulin production in the pig. Its object was to determine the proportion of IgA, IgG and IgM in sow colostrum and milk that is derived from serum by measuring the transfer of ^{125}I-labelled immunoglobulins from serum into colostrum and milk.

MATERIALS AND METHODS

Animals

Eighteen Large White sows, which had been mated to Large White boars, were used in this study.

Preparation of immunoglobulins

Serum IgG was prepared by the method of Curtis and Bourne (1971) and colostral and milk IgG were prepared by a similar technique (Curtis and Bourne, 1972). Serum, colostral and milk IgM were prepared by the method of Bourne (1971). The techniques used for the preparation of serum IgA and colostral and milk IgA are given elsewhere (Bourne, 1969a, b).

Iodination of immunoglobululins

Immunoglobulins were trace-labelled (approximately 5 μCi/mg) with ^{125}iodine by the Chloramine-T method of Hunter and Greenwood (1962) as described by Freeman (1966).

EXPERIMENTAL PROCEDURE

Serum-colostrum transport

Sows were injected intravenously with 30–120 mg of IgG, IgA or IgM labelled with ^{125}iodine, a week before the expected date of farrowing. At parturition colostrum was collected before suckling and a blood sample was taken. Immunoglobulin IgG, IgA or IgM was prepared from colostral whey and serum. The protein concentrations of the isolated immunoglobulins were measured by the radial immunodiffusion technique of Fahey and McKelvey (1965). Protein was precipitated from 4 ml of immunoglobulin solution with 10 per cent trichloracetic acid and the protein precipitates were counted in a gamma-ray counter (Panax Nucleonic Instruments). The specific activities (counts/mg) of immunoglobulin from serum and colostrum were compared and the ratio of these specific activities gave the proportion of immunoglobulin in colostrum which was of serum origin.

Serum-milk transport

Lactating sows were injected intravenously with ^{125}iodine-labelled immunoglobulin. A milk sample and a blood sample were collected 2 days later. Immunoglobulin was prepared from serum and milk whey and the specific activities of immunoglobulin from the two sources were measured and compared.

RESULTS AND DISCUSSION

The specific activities of IgG, IgA and IgM prepared from serum and colostrum or milk of sows injected intravenously with IgG, IgA or IgM labelled with ^{125}iodine are given in Tables 1, 2 and 3.

Colostrum

All colostral IgG and a high proportion of colostral IgM appear to be derived from serum. In contrast only 40 per cent of colostral IgA is of serum origin and this proportion falls rapidly with the onset of suckling. A sample of colostrum collected 6 hours after parturition contained only 11 per cent of IgA from serum.

Although 60 per cent of colostral IgA may be produced in the mammary gland, this is only a small fraction of the total since IgA constitutes only 13–15 per cent of colostral immunoglobulin (Porter, 1969; Curtis and Bourne, 1971). Thus it would appear that sow colostrum is a transudate and not a true secretion. This is similar to the situation in the cow and sheep where the major colostral immunoglobulin is fast IgG (Mach and Pahud,

TABLE 1

SPECIFIC ACTIVITY OF IgG (COUNTS/mg) PREPARED FROM THE SERUM AND COLOSTRUM OR MILK OF SOWS INJECTED WITH IgG LABELLED WITH ^{125}IODINE

Stage of lactation	Specific activity of IgG (c/mg) Serum	Specific activity of IgG (c/mg) Colostrum/milk	Ratio of c/mg serum IgG: c/mg col./milk IgG	IgG in colostrum or milk derived from serum (per cent)
Colostrum 0 hours	41	42	1:1·02	100
Colostrum 0 hours	150	160	1:1·07	100
Colostrum 0 hours	318	319	1:1·00	100
Milk 12 days	125	47	1:0·38	38
Milk 2 weeks	211	42	1:0·20	20
Milk 4 weeks	292	92	1:0·32	32
Milk 4 weeks	215	59	1:0·27	27

TABLE 2

SPECIFIC ACTIVITY OF IgA (COUNTS/MG) PREPARED FROM THE SERUM AND COLOSTRUM OR MILK OF SOWS INJECTED WITH IgA LABELLED WITH ^{125}IODINE

Stage of lactation	Specific activity of IgA (c/mg) Serum	Specific activity of IgA (c/mg) Colostrum/milk	Ratio of c/mg serum IgA: c/mg col./milk IgA	IgA in colostrum or milk derived from serum (per cent)
Colostrum 0 hours	38	9	1:0·24	24
Colostrum 0 hours	82	44	1:0·54	54
Colostrum 0 hours	650	255	1:0·39	39
Colostrum 6 hours	46	5	1:0·11	11
Milk 4 days	44	7·5	1:0·17	17
Milk 4 days	24	2	1:0·08	8
Milk 3 weeks	30	3	1:0·10	10
Milk 5 weeks	96	2	1:0·02	2

TABLE 3

SPECIFIC ACTIVITY OF IgM (COUNTS/MG) PREPARED FROM THE SERUM AND COLOSTRUM OR MILK OF SOWS INJECTED WITH IgM LABELLED WITH ^{125}IODINE

Stage of lactation	Specific activity of IgM (c/mg) Serum	Specific activity of IgM (c/mg) Colostrum/milk	Ratio of c/mg serum IgM: c/mg col./milk Igm	IgM in colostrum or milk derived from serum (per cent)
Colostrum 0 hours	39	30	1:0·77	77
Colostrum 0 hours	200	211	1:1·06	100
Colostrum 0 hours	151	106	1:0·70	70
Milk 4 days	25	8	1:0·32	32
Milk 3 weeks	1682	157	1:0·09	9
Milk 3 weeks	357	20	1:0·06	6
Milk 5 weeks	282	11	1:0·04	4

1971; Pahud and Mach, 1970) which appears to be actively and selectively transported from serum (Dixon, Weigle and Vasquez, 1961; Pierce and Feinstein, 1965). In all three species colostrum is the source of circulatory immunoglobulins to the newborn.

Milk

The results suggest that over 90 per cent of IgA and IgM and 70 per cent of IgG in milk is locally produced in mammary tissue. The transition from the colostral immunoglobulin profile in which IgG predominates to the immunoglobulin profile of milk in which IgA is the major immunoglobulin is complete a week after parturition in the sow (Curtis and Bourne, 1971). During this period the proportions of IgA and IgM derived from serum were intermediate between those of colostrum and milk. IgA accounts for 60 per cent of the total immunoglobulin in sows' milk and IgG and IgM 25 per cent and 15 per cent respectively (Curtis and Bourne, 1971). Therefore, local production accounts for 90 per cent of the immunoglobulin in milk.

The situation in sows' milk contrasts with that in bovine and ovine milk where fast IgG predominates and IgA is present only at very low levels (Mach and Pahud, 1971; Pahud and Mach, 1970). It has been suggested that the fast IgG in bovine and ovine milk is largely derived from serum (Dixon, Weigle and Vazquez, 1961; Mackenzie and Lascelles, 1968), although the evidence for this is inconclusive. After stimulation of sheep mammary gland with *Brucella abortus* antigen (Outteridge, Mackenzie and Lascelles, 1968), milk whey possessed detectable antibody activity associated with fast IgG and IgM. Most antibody activity was, however, associated with an immunoglobulin distinct from IgG and IgM, which was later provisionally identified as IgA (Lascelles and MacDowell, 1970). The latter authors proposed that this immunoglobulin was locally produced in response to antigenic stimulation.

In the rabbit, IgA is the major immunoglobulin of colostrum and milk, accounting for over 60 per cent of colostral immunoglobulin (Eddie *et al.*, 1971). These authors found that intravenous injection of heat-killed *Salmonella typhimurium* gave high bacterial agglutination titres in serum IgG and IgM of rabbits, accompanied by dramatic increases in the serum-levels of these immunoglobulins. However, colostrum agglutination titres were negligible and colostrum immunoglobulin levels were normal. Local immunization of the mammary gland with heat-killed bacteria gave rise to high levels of agglutinating activity in serum IgG and IgM but negligible levels of antibody in colostrum. However, local stimulation of the mammary gland with live organisms gave rise to antibodies in all three classes of immunoglobulin in colostrum and to increased levels of colostral IgM, IgG and IgA. These results indicate that all three classes of immunoglobulin are produced in rabbit mammary tissue. The authors suggested that the development of high levels of secretory antibodies may require persistent stimulation such as occurs by multiple administrations of a replicating antigen. Genco and Taubman (1969) compared the antibody types in colostrum of rabbits immunized with DNP-bovine gamma globulin by two routes; the foot pads and mammary tissue. IgG and IgM antibodies were found in the serum and colostrum of both groups, but IgA antibodies were only detected in the colostrum of rabbits immunized by the mammary route. Antibodies of the IgA class appeared to be locally produced in the mammary gland in response to this non-replicating antigen.

Lawton, Asofsky and Mage (1970) studied the *in vitro* production of IgA by rabbit mammary glands. ^{14}C-lysine and isoleucine were incorporated into IgA heavy (α), light and T chains in a ratio of approximately 4:2:1. They suggested that since this ratio was in reasonably close agreement with the mass-ratio of the chains of an IgA molecule with 4 α-4 light- and 2 T-chains, most if not all of the α- and light-chains were being synthesized locally and not just T-chains as had been previously suggested (Asofsky and Small, 1967).

In human colostrum antibodies to *Escherichia coli* were found to be associated mainly with IgA (Adinolfi, Glynn, Lindsay and Milne, 1966) which is the major immunoglobulin of this secretion. Antibodies of the IgA class could not be detected in the serum of the donors. They suggested that if the IgA antibodies in colostrum were derived from serum, the IgA content of colostrum ought to be at least a hundred times higher than that of serum. IgA levels in human serum and colostrum are in fact similar (Chodirker and Tomasi, 1963) and so IgA antibodies in colostrum must be synthesized in the mammary tissue.

The present study shows that milk IgA in the pig is almost entirely locally produced but that considerable amounts of IgG and IgM are also formed in mammary tissue, comprising 40 per cent of locally formed immunoglobulin.

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