

The IgE and IgG subclass antibody response to foods in babies during the first year of life and their relationship to feeding regimen and the development of food allergy

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This follow-up study of 191 babies investigated the development of food allergy in an unselected population and its relationship to total and antigen-specific IgE and IgG subclass levels. Sensitization to egg, as indicated by a positive skin test or RAST, was found in 5% of 1-year-old babies, but none of the babies in this series fulfilled the clinical criteria for immediate-type milk allergy. For both bovine casein (CAS) and egg albumin, the IgG response was largely restricted to IgG1 in contrast to the predominant IgG4 response to these antigens that is found in adults. The level of IgG4, but not IgG1, antibody to CAS and ovalbumin (OV) was lower in some of the babies compared with that of their mothers ($N = 166$; $p < 0.05$, Student's paired t test). However, there was no difference in the total serum IgG subclass levels between mothers and babies. These results demonstrate that, in the population of babies studied, (1) type I hypersensitivity to egg occurred in 5% of 1-year-old babies, (2) the predominant IgG subclass of antibodies to CAS and OV in babies is IgG1, and (3) in the 22% of babies, there was substantially (>1000 -fold) less IgG4 antibody to CAS and OV than in their mothers, suggesting specific exclusion of some IgG4 antibodies. (J ALLERGY CLIN IMMUNOL 1991;87:920-9.)

During the first year of life, the immune system is exposed to a wide range of foreign antigenic material to which an immune response may be made. Of this material, foods constitute a substantial antigenic load, and there are widely differing studies on the frequency with which infants become sensitized to food Ags.¹⁻⁴ Ingestion of foods has been associated with a brisk IgG, IgA, and sometimes IgE Ab response.⁴⁻⁷ A number of previous studies of the development of allergic disease in infants have concentrated on the ability to identify a baby's disease susceptibility by cord-blood IgE.^{8,9}

Little is known about immune responses in the different IgG subclasses during the first year of life. For

Abbreviations used

AP:	Alkaline phosphatase
PBS:	Phosphate-buffered saline
OV:	Ovalbumin
Ag:	Antigen
CAS:	Casein
Ab:	Antibody

infectious organisms, an appropriate response may be to produce cytotoxic or neutralizing Abs, whereas "self" Ags, foods, and some other environmental Ags may be tolerated. The Ab response to some Ags that are in themselves harmless can cause disease through the effects of IgE Abs in patients with atopic allergy or IgG in individuals with immune complex-mediated hypersensitivity. The subclass of IgG Ab differs for different Ags, and although IgE and immune complex-mediated reactions might be important in dealing with infectious organisms, such reactions would be inappropriate if they are provoked by food. The mechanisms that determine these inappropriate types of response are, however, poorly understood.

IgG Abs have rarely been studied in this early age

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Revised Nov. 27, 1990.

Accepted for publication Dec. 6, 1990.

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1/1/27353

group,² despite the evidence, in rats, that maternal IgG can inhibit IgE responses to foods.¹⁰ We have therefore studied the production of IgE and IgG Abs produced against common food Ags, milk, and egg in blood samples collected at 7 days, 3 months, and 1 year from a cohort of 191 babies born at Dulwich and King's College Hospitals in London in relation to total immunoglobulin levels, allergic disease, and feeding regimen.

MATERIAL AND METHODS

Subjects studied

Within 7 days of giving birth, 250 mothers at King's College and Dulwich Hospitals were invited to participate in the study; 191 (76%) agreed. After 1 year, 11 of these mothers had withdrawn from the study. All the babies were tested with common inhalant allergens by skin prick test with Bencard skin test solutions (Beecham, Betchworth, Surrey, U.K.) at 3 months and 1 year of age. A positive test was recorded when there was a wheal of 2 ml or more larger than the negative control wheal. This is a smaller size wheal than the generally accepted cutoff of 3 mm to reflect the smaller wheal reactions that are observed in babies. Judging from the correlation with serum IgE Abs and clinical symptoms, such a threshold appears to be justified in this age group. Blood samples were collected from all babies at 7 days and at 3 and 12 months and studied in a variety of ways. Of the serum samples collected, 191 of the 7-day samples, 161 of the 3-month samples, and 142 of the 1-year samples were available for the present study because some of the samples had been exhausted by assays for complement and because some mothers were unwilling for their babies to be bled. Clinical and skin test data were, however, available from 180 samples.

Symptoms

Wheezing on at least two separate occasions during the first year of life was used to define asthma and is generally referred to simply as wheeze. Eczema was defined according to the criteria described previously.¹¹ Symptoms of eczema, wheeze, or vomiting within 2 hours of eating the particular food on at least two occasions were taken to indicate a history of milk or egg allergy. Those babies in whom these symptoms occurred 2 to 24 hours after eating the food in question were regarded as possibly allergic, and in whom these symptoms occurred more than 24 hours later, as non-allergic.

Feeding

No additional instruction was provided to the mothers regarding the method of feeding other than that normally provided by the maternity services of these hospitals. The mothers freely chose the method of feeding that we then divided into breast-feeding only (for 3 or more months), mixed feeding when there had been some breast-feeding but cow's milk had been introduced before 3 months of age, and bottle feeding when the babies had never been breast-fed. No control of any kind was exerted on the mothers to determine the amount or type of feed used. The infants were

evaluated by the same clinician in a single-blind fashion. Clearly, the mothers knew how they had fed their infants.

Material

Nunc Immunlon-I microtiter plates were purchased from Gibco Ltd. (Paisley, Scotland, U.K.). Rabbit and horse serum were purchased from Sera Laboratories, Ltd. (Crawley, Sussex, U.K.). Tween 20, AP-labeled rabbit antimouse IgG, p-nitrophenyl phosphate, and human and mouse serum agarose were purchased from Sigma Ltd. (Poole, Dorset, U.K.). IgG subclass-specific mouse monoclonal Abs were purchased from Oxoid Ltd. (Bedford, U.K.). Monoclonal mouse antihuman IgE (clone 7.12) was a kind gift from Professor A. Saxon, University of California-Los Angeles. Ampak-amplified AP substrate was a kind gift from Dr. A. Johansson (Novo Bio Laboratories, Cambridge, U.K.). AP-labeled antihuman IgE and AP-labeled Fab' antihuman IgE were prepared as described previously.^{12, 13} Rabbit antimouse IgG and AP-labeled F(ab')₂ rabbit antihuman IgG were purchased from Dako (Copenhagen, Denmark). The latter was extensively absorbed with human-serum agarose and mouse-serum agarose.

Total IgE assay

All serum samples were diluted 1/20 in PBS, 0.05 mol/L, pH 7.4/0.5% Tween 20/0.5% normal rabbit serum and were first tested in a conventional two-site ELISA as described previously.¹² This method has a lower limit of detection of 0.5 IU/ml of IgE. Samples were assayed at a single dilution, in duplicate. Those samples with IgE levels too low to be detected with these methods were reassayed with a recently developed ultrasensitive ELISA¹³ that had a lower limit of detection of 0.01 IU/ml.

Conventional ELISA

All samples were assayed at a single concentration in duplicate and reassayed diluted if this were necessary. All volumes were 50 µl except for the substrate that was 100 µl, and all incubations except for the substrate (37° C) were performed at 4° C. Coating buffer was sodium carbonate/bicarbonate, 0.1 mol/L, pH 9.6; assay diluent was PBS/0.5% Tween 20/0.5% rabbit serum; substrate buffer was ethanolamine, pH 9.7, 0.1 mol/L; and wash solution was PBS/0.05% Tween 20. Briefly, Immulon-I microtiter plates were coated with mouse monoclonal antihuman IgE at 1 µg/ml in coating buffer overnight. After a wash step, the sample (diluted in the sample buffer) was added and incubated for 3 hours. The plate was then washed, and AP-antihuman IgE (diluted 1/500 in sample buffer) was added. After 3 hours, the plate was again washed, and the substrate was added and color was allowed to develop for ½ hours at 37° C. The optical density of the substrate was measured with an automated plate reader (Titertek Multiskan, Flow Ltd., Rickmansworth, Hertfordshire, U.K.) at 405 nm.

Ultrasensitive ELISA

ELISA was performed in the same way as the conventional assay with the following modifications: Tris HCl, pH

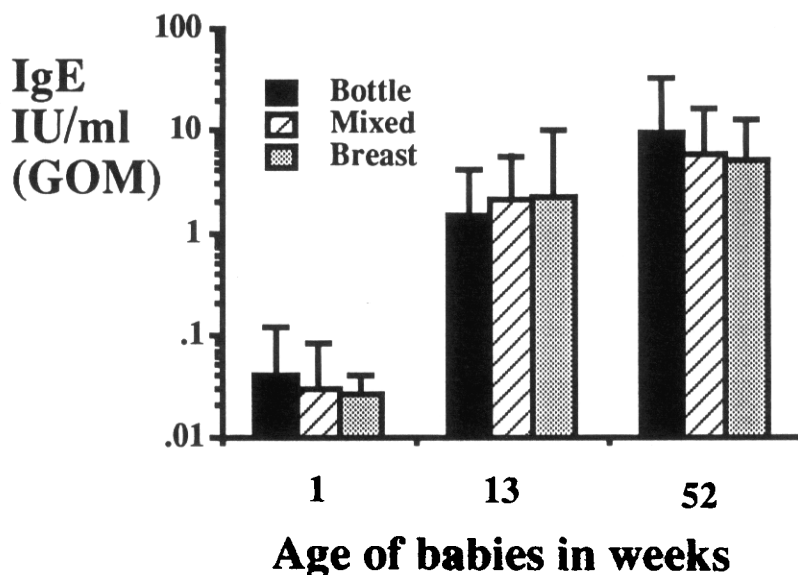


FIG. 1. Total serum IgE (geometric mean + SD) levels at 1, 13, and 52 weeks. The number of sera/patients analyzed here and in subsequent figures were 191, 161, and 142. The levels were not significantly different in babies fed by the different regimens indicated.

7.4, 0.1 mol/L, was substituted for the PBS, and AP-Fab' antihuman IgE at 1/3000 was substituted for the AP-antihuman IgE. An amplified substrate was used according to the manufacturer's instructions.

Total IgG subclass levels

Total IgG subclass protein was measured with a two-site ELISA. All assay volumes and buffers used were as described for the conventional IgE ELISA above. All samples were assayed at two dilutions in duplicate. Microtiter plates were coated with rabbit antimouse IgG at 1/3000 dilution. Mouse monoclonal anti-IgG subclass antibody (NL16, HP6014, ZG4, and RJ4) was added at 1/1000 dilution and allowed to bind for 1 hour. The plate was washed, and the sample at 1/1000 or greater dilution was added. After an incubation of 2 hours, AP-rabbit antihuman IgG (diluted 1/300) was added and incubated for 1 hour. Finally, the plates were washed, the substrate, *p*-nitrophenyl phosphate, 1 mg/ml, in pH 9.8 0.1 mol/L ethanolamine buffer, was added, and color was allowed to develop for 1½ hours. Color generated was measured at 405 nm as above. Results are expressed as micrograms per milliliter by reference to the World Health Organization/International Union of Immunological Societies reference preparation 67/97.

RAST

IgE Abs to milk and egg were measured by the micro-RAST.¹⁴ All samples were assayed in duplicate and all washes were with PBS/0.05% Tween 20 in a specially designed washing machine.¹⁵ Briefly, 5 µl of serum sample was added to a freshly washed allergen-coated disk. After a 10-minute incubation, the disk was washed and dried, and 5 µl of ¹²⁵I-labeled anti-IgE diluted in horse serum was added. After a 60-minute incubation, the disk was again washed and the bound radioactivity was counted in a gamma

counter (Multigamma, Pharmacia/LKB plc, Bromma, Sweden) for 1 minute. The results are expressed as a score from Phadebas RAST reference reagents.

Specific IgG subclass antibodies

IgG subclass antibodies to the major milk Ag, CAS, and egg Ag, OV, were measured by ELISA as described previously.¹⁶ Briefly, microtiter plates were coated with 3 µg/ml of OV or CAS (Sigma Ltd.). Samples were assayed at a single concentration, in duplicate. Each sample was diluted 1/50 or more in assay diluent, added to the plates, and incubated for 2 hours. Mouse monoclonal antihuman IgG subclasses 1 to 4 (NL16, Gom2, ZG4, or RJ4) were added in excess at a 1/1000 dilution. After a 1-hour incubation and washing, AP-antimouse IgG at 1/500 was added. After an additional 1-hour incubation and wash, the substrate was added and color was measured as above. There are no agreed standards for IgG Abs to foods or any other allergens. Weight estimates of IgG Ab can be made by radiolabeled Ag saturation analysis,¹⁶ but this analysis will only define the amount of Ab under the specific assay conditions used, or estimates by specific absorption and elution, which are prone to error.¹⁷ The presence of competing IgG subclasses for these Ags, and possible differences in affinity^{18, 19} and valency,²⁰ all make precise quantitation very difficult. We have therefore read from a standard curve generated by a series of dilutions of a positive serum pool with an IgG panreactive monoclonal (8a4) Ab as described in detail elsewhere²¹ and expressed these data as arbitrary units per milliliter.

Statistical analysis

The levels of OV and CAS-specific Abs were compared by the Mann-Whitney U test. Disease frequency was analyzed by the chi-square test, and paired samples were an-

TABLE I. The incidence of allergic symptoms and hypersensitivity to foods and their relationship to feeding regimen

	Symptoms		Allergy		RAST		Skin test	
	Eczema	Wheeze	Milk	Egg	Milk	Egg	Milk	Egg
Feeding regimen								
Bottle (N = 60)	10	14	0	1	0	2	0	2
Mixed (N = 64)	15	18	0	3*	0	2	1	3
Breast (N = 65)	11	8	0	1	0	4	1	4

A positive skin test was defined as a wheal of 2 mm larger than the negative control wheal. A positive RAST was a score of 1+ or higher.
*One of these allergies was probable egg allergy (case 10); all the other allergies were definite.

TABLE II. Clinical symptoms, skin test, total IgE, and RAST results in babies with a history of egg allergy

	Symptoms					RAST score (1 yr)	Skin test mm (1 yr)	Total IgE			Feeding regimen
	Wheeze	Eczema	Vomiting	Edema	Egg allergy			7 days	3 mo	1 yr	
Baby											
1	0	+	0	0	0	0	2	<	3.9	19	Bottle
2	0	0	0	0	0	1+	4	2.3	21	285	Breast
3	+	0	0	+	+	3+	4	<	13	4.8	Breast
4	0	0	+	0	+	1+	2	0.7	9.2	9.4	Mixed
5	0	+	0	0	0	2+	3	<	13	7.8	Breast
6	0	0	+	0	+	2+	3	<	19	4.3	Bottle
7	0	0	0	0	0	1+	0	ND	2	5.4	Bottle
8	0	0	0	0	0	1+	0	<	1	17	Breast
9	+	+	0	+	+	1+	3	1.2	36	290	Mixed
10	0	+	0	0	+	ND	2	<	21	68	Mixed
11	0	+	0	0	0	ND	5	<	100	134	Breast

+, Yes; 0, no; ND, not done; <, below 0.1 IU/ml.
The RAST and skin test results presented were performed at 1 year of age. These tests were performed at 3 months when tests were all negative.

alyzed from mother and baby by a two-tailed Student's paired *t* test. The interassay and intra-assay coefficient of variation of the different assays has been detailed elsewhere.^{12-14, 21} All tests were performed with Statview 512+ (BrainPower, Inc., Calabasas, Calif.) on an Apple Macintosh computer (Apple Computer, Inc., Cupertino, Calif.). A *p* value of <0.05 was accepted as statistically significant unless multiple comparisons were made. Under these circumstances, the *p* value required for statistical significance halved with each additional comparison.

RESULTS
Total serum IgE

Total serum IgE levels are illustrated in Fig. 1. These levels rose after birth and continued to rise between 3 months and 1 year. There was no difference in the IgE levels of babies fed with different regimens or in the babies who developed wheeze or eczema.

Allergic symptoms

None of the babies studied here had clear-cut evidence of a reaction to milk. All food-related symptoms were with egg. Four babies had symptoms within 2 hours of eating egg and one baby within 6 hours (Table I). The case histories of the five babies in whom eating egg was associated with allergic symptoms are discussed below. The case numbers provided refer to Table II. In the first four babies, a history of egg allergy was clear-cut, and it was not believed safe or ethically justifiable to perform formal challenge tests. The parents of these children, having witnessed the repeated adverse reactions, were reluctant for formal challenge tests to be done.

Case 3. This baby developed acute swelling of the face and generalized urticaria within 30 minutes of eating egg on two separate occasions. This infant also

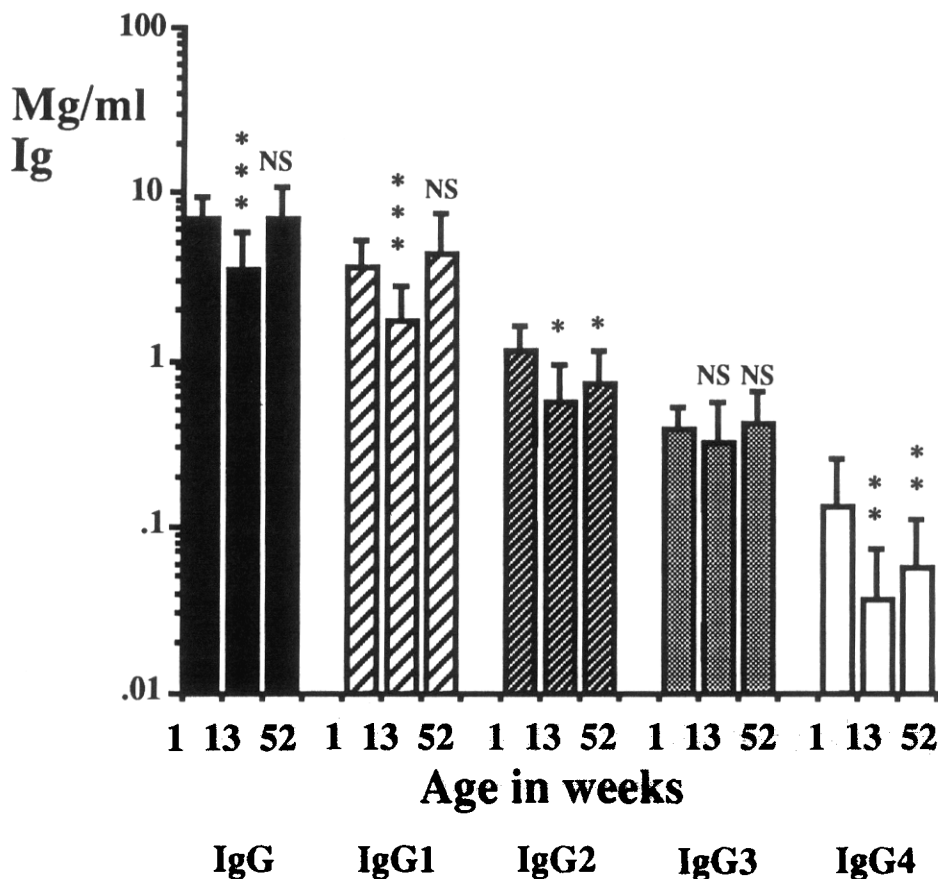


FIG. 2. Total IgG and IgG subclass (mean + SD) levels 1, 13, and 52 weeks. Total IgG fell between 1 and 13 weeks ($p < 0.001$) but regained the level observed at 1 week by the end of the year. IgG subclasses 1, 2, and 4, but not 3, fell between 1 and 13 weeks. IgG2 and IgG4 failed to reach the 1-week level by 52 weeks; *** $p < 0.0001$; ** $p < 0.01$; $p < 0.025$, Student's paired t test.

had at least two episodes of wheezing, but these were not associated with ingestion of egg.

Cases 4 and 6. Thirty minutes after eating egg, these babies developed acute vomiting. This occurred on three occasions. There were no other manifestations of atopy.

Case 9. On three separate occasions this baby developed acute swelling of the face and generalized urticaria within 30 minutes of eating egg. This infant also had eczema and recurrent episodes of wheezing that were not specifically associated with eating egg.

Case 10. Within 6 hours of eating egg, this infant developed an acute exacerbation of eczema with erythema, pruritus, and vesicle formation. No formal challenge test was performed because of parental unwillingness.

IgE Abs to milk and egg

Sera were assayed for the presence of IgE Abs in the 3-month and 1-year samples. No IgE Abs could

be detected against milk in any of these or against hen's egg in any of the 3-month samples. Eight (5.6%) of the 142 1-year samples tested contained IgE Abs to hen's egg, and nine of 180 babies who were skin tested were positive (5%). Six of the eight egg RAST-positive babies also had positive skin prick tests with egg. Egg skin tests, total IgE, RAST scores, and clinical symptoms are presented in Table II. It is interesting to note that 9/11 of these babies were wholly or partly breast-fed for the first 3 months of life.

Total serum IgG subclasses 1 to 4

Total serum IgG subclass levels were measured at 7 days, 3 months, and 1 year. The levels of IgG1, IgG2, and IgG4, but not IgG3, fell between 7 days and 3 months (Fig. 2). IgG1 and IgG3 levels returned to the levels measured at 7 days by 1 year, whereas IgG2 and IgG4 levels were still lower than at birth (Student's paired t test IgG2, $p < 0.025$; IgG4, $p < 0.01$).

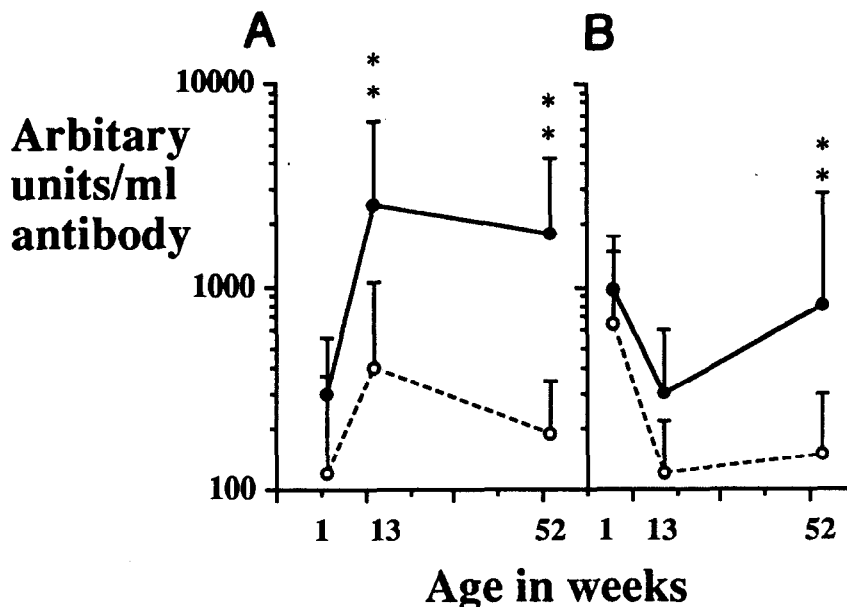


FIG. 3. The IgG1 (●—●) and IgG4 (○- -○) antibody (mean + SD) response. **A,** To CAS. **B,** To OV. Significantly more IgG1 than IgG4 antibody to CAS was detected at 13 and 52 weeks and to OV at 52 weeks ($p < 0.001$, Mann-Whitney U test).

IgG subclass Abs to CAS and OV

IgG subclass Abs to CAS and OV were measured in the serum samples collected from the babies at 7 days and 3 and 12 months. For both Ags, most Ab detected was in IgG subclasses 1 and 4 (Fig. 3, A and B). CAS-specific Ab levels rose between 7 days and 3 months ($p < 0.001$, Mann-Whitney U test). At 12 months, the levels reached a plateau and were not statistically different to levels measured at 3 months (Fig. 3, A). In contrast, the levels of OV-specific Abs fell between 7 days and 3 months ($p < 0.001$, Mann-Whitney U test) but had risen by 12 months ($p < 0.005$, Mann-Whitney U test), especially IgG1 (Fig. 3, B). For both Ags, most of the antibody was IgG1.

The relationship between feeding regimen and the IgG subclass Ab response

The relationship between the amount and subclass of IgG Ab to CAS and OV was also analyzed according to the feeding regimen used. Babies were divided into babies who had been exclusively breast-fed for the first 3 months, babies exclusively bottle-fed, and babies who had had a mixture of breast- and bottle-feeding. The results (Fig. 4) demonstrated that the bottle-fed babies made the most vigorous IgG Ab response to CAS and that the breast-fed babies made the weakest response. By 12 months, the levels were

falling in the bottle-fed babies but still rising in babies who were breast-fed.

There was no evidence that feeding had any effect on the levels of OV-specific Ab. These levels were initially at or below the levels found in maternal serum samples but fell between 7 days and 3 months (presumably as a result of the loss of maternal-derived Ab) and had risen by 12 months.

Transplacental transport of IgG subclass proteins

When paired samples from mother and baby (7-day) were compared (Fig. 5, A and B), there was no statistically significant difference between the levels of maternal and infant IgG1 Abs to CAS and OV. The levels of IgG4 Abs to these Ags were, however, lower in the babies than in their mothers ($p < 0.05$; $N = 166$; Student's paired t test). In 36 paired samples, the discrepancy between maternal IgG4 Ab and the baby's IgG4 antibody was considerable (>3000 U/ml). To determine if this was due to a selective deficiency in transplacental transport of IgG4, the levels of IgG subclasses 1 to 4 in these 7-day serum samples and samples from the corresponding mothers were measured (Fig. 6). There was no statistically significant difference between mother and baby in the levels of any of the IgG subclasses. Poor transport of IgG4 (or of IgG proteins in general) cannot therefore

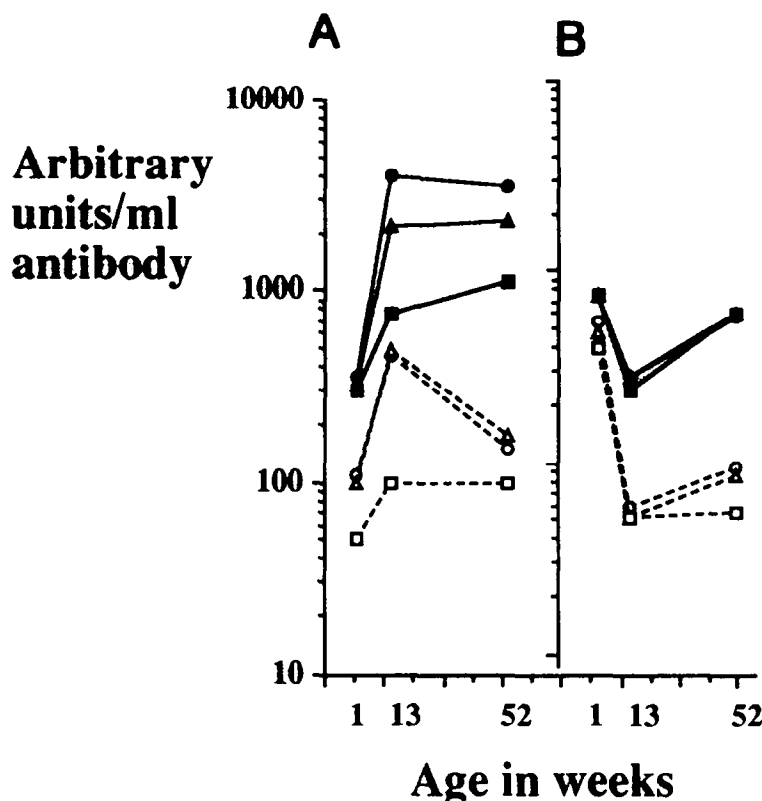


FIG. 4. The mean levels of IgG1 (—) and IgG4 (---) antibody. **A,** To CAS. **B,** To OV. In breast-fed (■) mixed-fed (▲), and bottle-fed babies (●). The level of IgG1 antibody to CAS at 3 months was greater in the bottle-fed babies compared with that of babies who were partially or exclusively breast-fed ($p < 0.01$ and $p < 0.001$, respectively; Mann-Whitney U test).

explain the difference in levels between mothers and babies at 7 days.

DISCUSSION

The purpose of this study was to investigate the development of food allergy during the first year of life in an unselected population and to identify the subclass of IgG Ab produced on first contact with food Ags. The prevalence of cow's milk allergy may be declining. When we compared studies of a high frequency of milk allergy described by other investigators²² with our study, we found no babies in this series who had clear-cut clinical reactions to milk or who had milk-specific IgE Abs. Evidence of sensitization to egg (a positive skin test or RAST), in contrast, was observed in 11 babies at 1 year of age. This finding was supported in four cases by the rapid onset of allergic symptoms associated with eating egg and in one case by delayed symptoms (6 hours), in three cases with wheeze or eczema not directly associated with eating egg, and in only three cases with a complete absence of clinical symptoms.

Although there is evidence to suggest that cord-blood total IgE levels can identify babies at risk of

developing allergy later in life,^{8,9} we found that many of the babies who developed egg allergy did not have a raised cord-blood IgE. Previous studies on the significance of serum IgE levels at 3 or 4 and 12 months are divided. Zeiger et al.²³ reported that IgE levels were raised at 4 and 12 months in babies who had atopic dermatitis or urticaria, but not in babies who had asthma. Matthew et al.¹¹ demonstrated that IgE levels were raised in babies with eczema at 3 and 12 months but not at 6 weeks and 6 months, and Hattevig et al.²⁴ did not find any difference in serum IgE levels in atopic and nonatopic high-risk babies. In this study, we found that IgE levels at 3 and 12 months were not significantly higher in those babies who had eczema or wheeze but were higher in babies who developed egg allergy. Feeding regimen had no effect on the level of serum IgE.

Previous studies of the subclass of IgG produced during a specific immune response have demonstrated that this can be influenced both by the type of Ag²⁵ and by the degree of antigenic stimulation.^{26,27} Our present data demonstrate that even for Ags like OV, which produce an IgG4-restricted response in adults, induce predominantly IgG1 antibodies in babies.

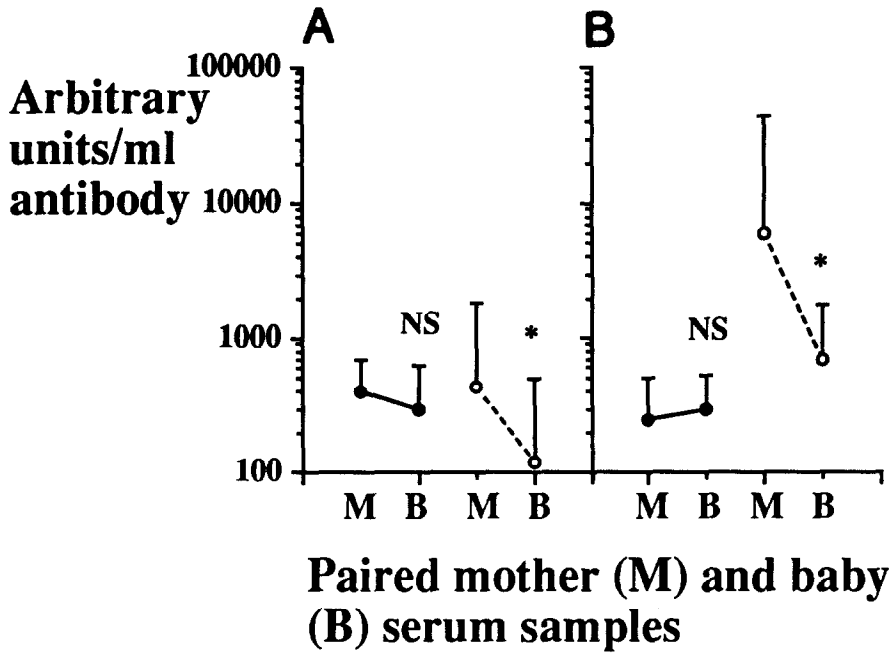


FIG. 5. Comparison of the IgG1 and IgG4 antibody levels. **A**, Against CAS. **B**, Against OV. In 166 paired samples from mother and baby collected 7 days post partum; the mean + SD are illustrated. IgG4 (○- - -○) but not IgG1 (●—●) antibody levels were significantly lower in the babies compared with the levels in their mothers ($p < 0.05$, Student's paired t test).

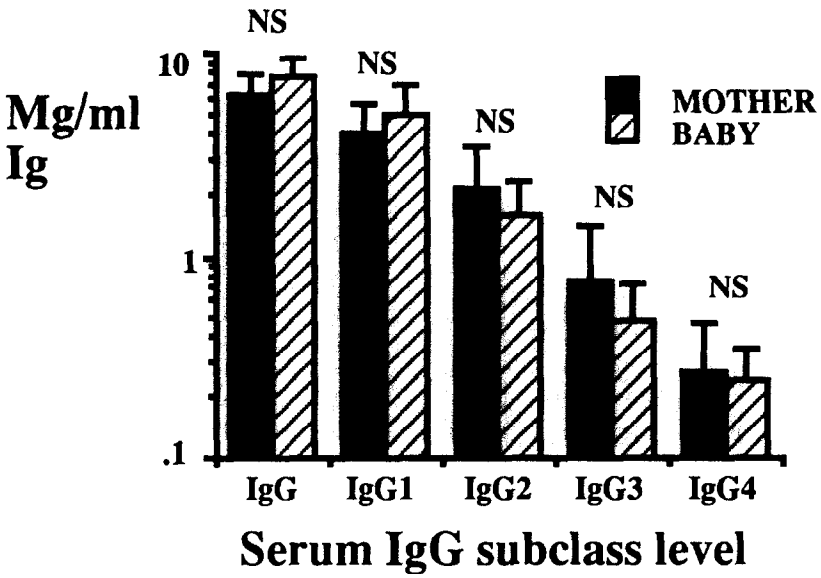


FIG. 6. Comparison of total serum IgG subclass levels in 36 paired serum samples from mothers and babies in which the IgG4 anti-OV or CAS antibody levels in the babies were >1000 -fold lower than levels present in the mothers. There was no statistically significant difference in the levels of any of the IgG subclasses between mother and baby.

Thus, the subclass of IgG produced early in the immune response to Ag, whether by injection,²⁸ via the gut,²⁵ or by inhalation,²⁹ is largely confined to subclass 1. As might be expected from the time at which they

are introduced into the diet, the antibody response to milk CAS preceded the response to OV whose pattern reflected more closely total IgG levels that fell between birth and 3 months and regained their birth levels by

1 year (with the exception of IgG2 and IgG4). Bottle-fed babies produced high levels of IgG Ab to CAS earlier than babies who followed a mixed or breast-feeding regimen, although the levels of IgG Ab in these babies appeared to be reaching the levels of their bottle-fed counterparts by the time 1 year was reached. Other studies on the effect of diet on IgG Ab responses to cow's milk reveal conflicting results. Eastham et al.²⁹ demonstrated that babies receiving soy milk and CAS hydrolysate had significant IgG responses to milk whey during the first 3 months, whereas Fåltch-Magnusson et al.⁷ demonstrated a fall in IgG Ab to β -lactoglobulin during the first 3 months of life and Hattevig et al.²⁴ was unable to detect IgG Ab to β -lactoglobulin at birth. These differences may be due, in part, to the methods used (hemagglutination and ELISA) and the Ags involved (β -lactoglobulin, milk whey, and CAS). It is also possible that some unreported feeding of the breast-fed babies with cow's milk may have occurred.

Total IgG subclass levels were measured in a representative group of babies at 7 days and 3 and 12 months. It has been reported in some studies that IgG1 levels are higher and IgG2 levels are lower in fetal than in maternal sera.³⁰⁻³³ This is, however, not a universal finding.³⁴⁻³⁶ Although the level of IgG1 was higher and IgG2 was lower in our 7-day samples as compared with matched maternal sera, the differences observed did not achieve statistical significance. Between 7 days and 3 months, the levels of IgG1, IgG2, and IgG4, but not IgG3, fell. The early activation of IgG3 responses could be a consequence of early viral infection, and this is currently being investigated in a specially selected population of babies.

An unexpected finding was that in approximately a fourth of the mother and baby pairs,³⁶ the IgG4 antibody level was at least 1000 times greater in the mother than in the baby. In these 36 cases, comparable total IgG-subclass protein levels were found in paired sera from mother and baby. Defective transport across the placenta appears therefore an unlikely explanation. It is possible, however, that a preferential tendency for IgG4 Abs to form immune complexes with ingested food might have played a part by making maternal Ab less available for placental transport. It is not clear why one subclass of Ab might form immune complexes in preference to another. Recent studies of bee venom-allergic children receiving immunotherapy have demonstrated that the affinity of IgG4 Abs produced after prolonged immunization is approximately 100-fold higher than that of corresponding IgG1 Abs.¹⁸ Furthermore, similar differences have also been observed for IgG4 compared with IgG1 Abs to OV and CAS.¹⁹ It is therefore possible that maternal

food ingestion could provide sufficient circulating food protein to form complexes with maternal IgG antibody. The amounts of food crossing the gut have been demonstrated to be in the order of 1 to 5 ng/ml.³⁴ Such small quantities could be expected to complex preferentially with high-affinity Ab (IgG4). The transport of complexed Ab across the placenta might well be less than that for free Ab and explain the relative paucity of IgG4 Abs in some babies.

This study demonstrated that, in the population studied here, milk allergy was rare compared with egg allergy and that total serum IgE levels at 7 days cannot be used to predict the development of egg allergy. Additional studies of babies who are at risk of developing allergy are in progress, and it is hoped that these studies will provide more information about the pathogenesis of food allergy in early life.

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