

Incremental prognostic factors associated with cow's milk allergy outcomes in infant and child referrals: the Milan Cow's Milk Allergy Cohort study

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Background: The prognosis for many children with cow's milk allergy (CMA) is remission within 3 years, and the clinical parameters that predict duration of disease have not been measured incrementally.

Objective: To prospectively determine prognostic predictors of tolerance in a random cohort of referrals using CMA workup outcomes as covariates and tolerance as the status variable in a duration model of CMA.

Methods: The 2001–2006 Milan Cow's Milk Allergy Cohort (MiCMAC) enrolled children referrals using double-blind, placebo-controlled food challenges (DBPCFCs) as study end points (confirmation of CMA; onset of tolerance). The Cox regression model was used to analyze all clinical factors that contributed to tolerance. Covariates analyzed were skin, gastrointestinal, and respiratory symptoms; history and demographics at presentation; age at diagnosis and DBPCFC outcomes; sensitization (skin and serum) by cow's milk protein fractions; sensitization to other food and inhalant allergens; total IgE levels; specific IgE concentrations for cow's milk protein fractions, other ingestants, and aeroallergens; and threshold doses at DBPCFC. Sensitization and DBPCFC were performed at 6-month intervals.

Results: A total of 112 infants were enrolled (mean [SD] age, 13.85 [9.84] months), and 59 achieved tolerance (mean [SD] age when tolerance was achieved, 27.58 [11.81] months). On univariate analysis, asthma and/or rhinitis at presentation was an independent predictor of persistence (hazard ratio [HR], 2.19; 95% confidence interval [CI], 1.26–3.82). On multivariate analysis, predictors of persistence were a fresh milk wheal diameter increment of 1 mm (HR, 1.18; 95% CI, 1.07–1.31) and a positive skin prick test result with soy (HR, 6.99; 95% CI, 1.56–31.25).

Conclusions: This is the first study, to our knowledge, to identify incremental biological predictors of delayed tolerance to cow's milk in children that should be integrated into DBPCFC schedules for CMA in infants.

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INTRODUCTION

A gap in the literature exists regarding the modeling of the duration of cow's milk allergy (CMA) and the acquisition of tolerance. In general, a reported 50% of infants see their cow's milk-induced symptoms disappear before their first birthday¹ and most children achieve tolerance before their fourth year,^{2,3} but the natural history of tolerance remains to be written for populations of referred patients. The most reliable data on prognosis are drawn from birth cohorts^{1,4} but are limited for selected populations.^{2,3,5} Only 2 studies^{2,5} used the double-blinded, placebo-controlled food challenge (DBPCFC) as the reference standard for diagnosis, whereas some researchers analyzed IgE-mediated and non-IgE-mediated reactions together.^{1,4–7} The age when the initial symp-

toms develop is not always reported in studies of CMA referrals,^{4,8} whereas some designs are retrospective.^{5,9}

Symptom severity after cow's milk ingestion,⁶ large wheal response to skin prick tests (SPTs), and a high specific IgE antibody titer to bovine proteins^{1,4} have thus been described as prognostic factors in CMA, but which clinical parameters actually contribute to the duration of CMA and by what order of magnitude remain unclear in pediatrics. A negative IgE test result at diagnosis¹⁰ or a 99% decrease in specific IgE levels during more than 12 months⁵ have been said to be predictive of tolerance. Other factors, such as family history, comorbidities, and presenting symptoms, have still not been investigated prospectively in the context of tolerance acquisition among children with CMA. A lack of consistency also exists across studies regarding the association between duration of disease and clinical parameters, such as immediate symptom severity, large skin prick wheal diameter, elevated specific IgE antibody titer to cow's milk proteins, and decreased specific IgE levels during 12 months.^{2,4,6,7,9–11} In the present study, we prospectively recruited a cohort of infants and children (referred to as the Milan Cow's Milk Allergy Cohort [MiCMAC]) with IgE-mediated CMA confirmed by

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DBPCFC admitted to our tertiary care allergy unit and clinically followed up using DBPCFC-evaluated tolerance as the observation end point. We planned a 2-pronged study. In the clinical arm of MiCMAC, we focused on the patient variables that affect tolerance achievement in a duration model of CMA among pediatric referrals. In an investigative arm of MiCMAC, we studied the quantitative variations of allergy parameters as independent predictors of persistence of disease in a duration model of CMA and tolerance. Several authors have found a threshold value for parameters for the degree of allergen sensitivity (SPT wheal diameter and specific IgE titers), but, to our knowledge, this is the first prospective evaluation of incremental measurements in connection with duration of disease.

METHODS

The MiCMAC Study

Our institution is a regional referral center for the prevention, diagnosis, and therapy of pediatric allergic disease. In planning the MiCMAC study, we defined observation as the period between a positive diagnostic DBPCFC procedure and a subsequent negative DBPCFC, confirming the acquisition of tolerance. Infants and children referred for suspected CMA evaluation between January 1, 2001, and December 31, 2005, were considered for the MiCMAC study if positive to DBPCFC with cow's milk as part of their workup. Informed consent was obtained for oral food challenge procedures. Nutritional, allergy, and growth evaluations were also included.

Study Design

This study was designed to prospectively determine incremental prognostic predictors of tolerance in a random cohort of referrals using CMA workup outcomes as covariates and tolerance as the status variable in a duration model of CMA.

Definitions

Family history of atopic manifestation, including food allergy, was recorded. Family history was defined as 1 parent or sibling with a physician's diagnosis of allergic rhinitis, asthma, anaphylaxis, and/or atopic eczema.¹² History of food allergy was defined as 1 parent or sibling with physician-diagnosed food allergy (by means of SPT, specific IgE determinations, oral food challenge, or a combination thereof). Patient risk factors (reported by parents or based on patient medical records) were as follows: eczema; urticaria and/or angioedema; anaphylaxis; gastrointestinal symptoms; respiratory symptoms (asthma and/or rhinitis); familiarity for allergic disease; sex; birth weight and length *z* score; age at onset of symptoms; age at first oral food challenge procedure; sensitization to cow's milk expressed as a SPT wheal diameter larger than 3 mm and a specific IgE level above 0.35 kUI/L; cosensitization to other inhalant and food allergens (defined as at least 1 positive SPT result and/or specific IgE determination); and dose eliciting a positive oral food challenge with cow's milk. Patient history of atopic eczema,

urticaria, angioedema, asthma, gastrointestinal symptoms, and anaphylaxis were recorded according to parents' report and clinical medical records when available. Eczema was defined following current guidelines.¹³ Urticaria and/or angioedema was defined as a generalized or local vascular reaction of the skin with erythema and wheal formation after ingestion of milk.¹³ Asthma was defined as 3 separate episodes of wheezing in a clinical setting where asthma is likely and other conditions have been excluded, 3 separate episodes of nocturnal cough with sleep disturbances for at least 3 consecutive nights, or a combination of 3 such episodes.¹⁴ Rhinitis was defined as a symptomatic disorder of the nose induced by an IgE-mediated inflammation after allergen exposure of the membranes lining the nose.¹⁵ The definition of anaphylaxis was "a severe, life-threatening, generalized or systemic hypersensitivity reaction."¹³ We defined gastrointestinal symptoms as vomiting and/or diarrhea that occurred within 30 minutes of ingestion of foods for which there was a positive sensitization record.¹⁶

Clinical Evaluation and Patient Variables

Pediatric referral patients with suspected immediate reactions to cow's milk were followed up according to our institutional workup protocol. An allergy, dietary, nutritional, and growth status evaluation was performed. All children presented with a convincing history of immediate CMA, and sensitization was evaluated by either a positive SPT or blood test result. When confirmed allergic at DBPCFC, children were prescribed a cow's milk-free diet. If infants were breastfed, breastfeeding was encouraged and a maternal milk-free diet was recommended. Symptomatic, nonbreastfed infants were randomly allocated to a partial or total supplementation diet, and a cow's milk extensive hydrolysate, a rice hydrolysate, and a soy formula were offered to substitute cow's milk at 4-month intervals. However, the use of prescribed cow's milk substitutes on which children fared well before enrollment was not discontinued. Follow-up visits were planned on a 3-month basis (± 14 days). Sensitization to food allergens was evaluated every 6 months by SPT and specific IgE determinations. Follow-up DBPCFCs were scheduled at 6-month intervals for 18 months and yearly thereafter. DBPCFCs were anticipated if inadvertent cow's milk ingestion without symptoms was reported. If unintentional ingestion induced clinical symptoms, the ingested reported dose was recorded and the next scheduled DBPCFC was delayed accordingly. Inhalant allergen sensitization was evaluated by SPT only. The nutritional and growth status of patients and weight-for-age and length-for-age *z* scores were also evaluated. Duration of disease was defined as time between a confirmed diagnosis of CMA at oral food challenge and the first negative oral food challenge, both performed under double-blinded conditions.

Skin Prick Testing

SPTs for ingestant (milk, egg white and yolk, beef, codfish, rice, soy, wheat, and peanut) and inhalant (grass mix, *Der-*

matophagoides pteronyssinus, *Dermatophagoides farinae*, and dog and cat dander) allergens were performed by standard technique. A solution of histamine phosphate in 50% glycosaline (10 mg/mL) and the diluent were used as positive and negative controls, respectively. Wheal diameters were read through a clear plastic caliper disk scaled in quarter-millimeters less than $\times 4$ magnification and recorded as positive when a 3-mm or larger wheal margin (SPT cutoff point) was included within a complete caliper circle, rounding measurements to the nearest millimeter.¹⁷ SPTs with fresh or commercial beef reagents were planned as were SPTs with fresh, pasteurized milk and, whenever possible, commercial milk tests (cSPT; ALK-Abellø, Copenhagen, Denmark). Because of safety concerns related to the bovine spongiform encephalopathy crisis in Europe, commercial reagents for cow's milk and beef were unavailable for part of the lifetime of the study and were only reintroduced in April 2006. Thus, from April 2002 onward, commercial cow's milk allergens for SPTs were substituted by at least 2 of the following reagents: α -lactalbumin, β -lactoglobulin, and total casein (all from Sigma Chemical Co, St Louis, Missouri). These allergens were solubilized at 5% final protein concentration. SPTs were not performed in children with severe eczema and/or when antihistamine medication could not be suspended.

Specific IgE Determinations

Serum concentrations of IgE to cow's milk, α -lactalbumin, β -lactoglobulin, casein, beef, egg white, egg yolk, codfish, soy, wheat, and rice were determined by processing patients' serum samples from venous blood samples obtained at the time of oral food challenges with the automated Pharmacia CAP system FEIA (Pharmacia & Upjohn Diagnostics, Uppsala, Sweden). The cutoff point for positivity was set at 0.35 kU/L.

Oral Food Challenges

DBPCFCs were performed according to Bock's method¹⁸ and blinded according to the European Academy of Allergy and Clinical Immunology position paper.¹⁹ Fresh, pasteurized cow's milk was administered in incremental doses of 0.1, 0.3, 1.0, 3.0, 10, 30, and 100 mL at 30-minute intervals for 3½ hours, using Neocate (Numico, Liverpool, England) as placebo. Neocate was added to the verum in a standardized manner to even out differences in taste. The placebo sequence was administered in identical doses on a separate day. Oral food challenge was discontinued at the first onset of symptoms or if no symptom developed after the seventh dose. For the purpose of the study, only reactions to DBPCFCs occurring within 6 hours are reported.

Statistical Analysis (Duration Model)

The median and the quartiles of CMA duration were estimated by Kaplan-Meier survival analysis. The association of risk factors with disease duration was evaluated by a univariate proportional hazard Cox regression model for continuous and categorical variables. Because in this analysis negative

prognostic factors generate hazard ratios (HRs) below 1, we report inverse HR with confidence intervals (CIs). To identify the independent predictors of the duration of the condition, a multivariate Cox model with stepwise selection was used, including all the variables significantly associated with survival at univariate analysis. Because of their markedly skewed distribution, total and specific IgE were log-transformed before analysis. $P < .05$ was the significance level set, and all tests were 2-tailed. Thus, HRs were calculated for each increment of 1 mm at SPTs and as a 1-log increment in specific IgE titer, total IgE concentration, and DBPCFC dose. All analyses were performed using the SAS statistical package version 8.2 (SAS Institute Inc, Cary, North Carolina).

RESULTS

The MiCMAC Study

Between 2001 and 2005, 339 infants and children were referred to our institution for evaluation of their immediate-onset symptoms and CMA workup (Fig 1). A total of 192 patients (56.6%) were confirmed as being allergic to cow's milk by DBPCFCs. A total of 147 procedures produced negative results. Thirty-nine patients did not complete the protocol because they were transferred to other cities ($n =$

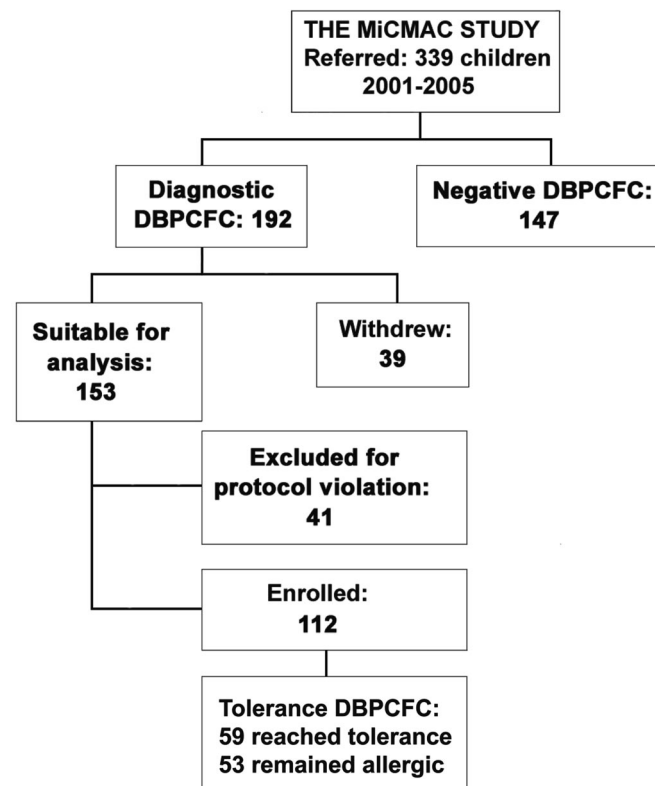


Figure 1. The Milan Cow's Milk Allergy Cohort (MiCMAC) patient flowchart. DBPCFC indicates double-blind, placebo-controlled food challenge.

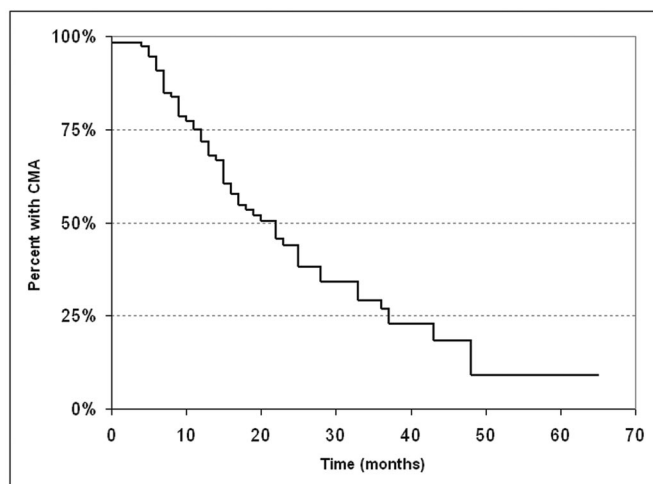


Figure 2. The Milan Cow's Milk Allergy Cohort survival curve for onset of tolerance to cow's milk allergy (CMA) (Kaplan-Meier analysis; percentage of patients surviving for each interval).

11), refused to undergo subsequent DBPCFCs ($n = 20$), or were lost to follow-up ($n = 8$); thus, 153 patients were considered suitable for analysis. Forty-one children were not included because of protocol violations (failure to comply with oral food challenge schedule, serum test or SPT refusal, severe disease unrelated to allergy). The only significant difference between children not included and the studied population was the frequency of urticaria at presentation (47% vs 67%; $P = .04$). All other parameters considered did not differ among these populations.

Thus, 112 infants (39 girls and 73 boys; mean [SD] age, 13.85 [9.84] months; median age, 10.5 months; age range, 1–61 months at diagnostic DBPCFC) completed the study and were followed up for a mean (SD) period of 18.15 (12.71) months (range, 3–60 months; median, 14 months).

Fifty-nine (52.7%) eventually reached tolerance evaluated by DBPCFCs. The median duration of disease was 23 months from the diagnostic challenge within a period of 4 to 56 months. Figure 2 shows the survival curve for tolerance in the MiCMAC study. Mean (SD) age at DBPCFC-evaluated tolerance was 27.58 (11.81) months (median age, 24 months; age range, 9–59 months). A total of 25% of children achieved tolerance within 13 months from diagnostic DBPCFC (95% CI, 10–16 months), 50% within 23 months (95% CI, 17–28 months), and 75% within 43 months (95% CI, 33–56 months). Patients who did not reach tolerance were observed up to a mean (SD) age of 34.73 (15.88) months (median age, 34 months; age range, 6–60 months). The mean (SD) wheal diameter of SPT with fresh milk was 6.43 (2.62) cm (range, 0–14 cm) among patients reaching tolerance and 8.87 (3.40) cm (range, 0–18 cm) among patients still not tolerant at the end of observation. Mean (SD) milk specific IgE concentrations were 6.25 (9.88) kUI/L (range, 0–54.12 kUI/L) and 20.76 (27.09) kUI/L (range, 0–100 kUI/L) among tolerant and nontolerant children, respectively. Mean (SD) total serum IgE concentration reached 149.86 (288.61) kUI/L (range, 6.07–6,885 kUI/L) in the group of children tolerant of milk within the study limits and 210.17 (316.03) kUI/L (range, 2–2,288 kUI/L) among those who had not achieved tolerance.

Univariate Analysis for Independent Predictors of Tolerance

The clinical features factored in the univariate analysis were stratified according to tolerance achievement and are reported in Tables 1 to 3. Table 1 lists the MiCMAC study demographics and the symptoms that prompted referral. Reported symptoms developed at a mean (SD) age of 2.62 (1.69) months (median, 2.0 months). At univariate analysis, asthma and/or rhinitis at presentation generated an HR of 2.19 (95% CI, 1.26–3.82; $P = .005$) and were the only statistically significant clinical history factors associated with failure to reach tolerance within our observation.

Table 1. Demographic and Clinical Variables Assessed by Univariate Cox Analysis^a

Factor	Patients reaching tolerance (n = 59)	Patients not reaching tolerance (n = 53)
Patient demographics		
Age at referral, mean (SD), mo	11.68 (7.64)	16.26 (11.41)
Male, No. (%)	41 (69.49)	32 (60.37)
Age at onset of presenting symptoms, mean (SD), mo	2.86 (1.84)	2.36 (1.48)
Age at diagnostic DBPCFC, mean (SD), mo	10.37 (6.44)	14.72 (9.79)
Symptoms at presentation, No. (%)		
Atopic eczema and/or dermatitis syndrome	45 (76.27)	48 (90.56)
Urticaria and/or angioedema	39 (66.10)	37 (69.81)
Asthma and/or rhinitis ^b	20 (33.90)	23 (43.39)
Gastrointestinal symptoms	13 (22.03)	14 (26.41)
Anaphylaxis	4 (6.78)	9 (16.98)

Abbreviation: DBPCFC, double-blind, placebo-controlled food challenge.

^a The total number of patients is more than 100 because of multiple presenting symptoms.

^b The inverse hazard ratio (95% confidence interval) for asthma or rhinitis was 2.19 (1.26–3.82). This finding was the only statistically significant result ($P = .005$).

Table 2. The MiCMAC Sensitization Variables (Skin Prick Test Wheal Diameters and sIgE Concentrations) Stratified by Tolerance Outcome

Factor	Patients reaching tolerance (n = 59)	Patients not reaching tolerance (n = 53)
Bovine allergen positive (>3-mm wheal diameter) skin prick test results, No. (%)		
Cow's milk reagent	37/41 (90)	19/23 (83)
α -Lactalbumin	21/30 (70)	18/25 (72)
β -Lactoglobulin	19/23 (83)	16/21 (76)
Casein	40/47 (85)	22/27 (81)
Fresh cow's milk	53/59 (90)	46/53 (87)
Beef	6/25 (24)	5/11 (45)
Bovine allergen sIgE concentrations >0.35 kUI/L in the 0.35–100 range of the assay, No. (%)		
Cow's milk	51/59 (86)	46/53 (87)
α -Lactalbumin	43/59 (73)	38/53 (72)
β -Lactoglobulin	36/59 (61)	37/53 (70)
Casein	45/59 (76)	44/53 (83)
Beef	18/55 (33)	21/40 (52)
Nonmilk food allergen positive (>3-mm wheal diameter) skin prick test results, No. (%)		
Egg white	34/58 (59)	32/42 (76)
Egg yolk	31/58 (53)	32/42 (76)
Rice	3/49 (6)	6/37 (16)
Wheat	2/52 (4)	12/38 (32)
Peanut	7/44 (16)	13/43 (30)
Codfish	3/48 (6)	4/37 (11)
Soy	2/59 (3)	6/38 (16)
Food allergen sIgE concentrations >0.35 kUI/L in the 0.35–100 range of the assay, No. (%)		
Egg white	32/54 (59)	39/49 (80)
Egg yolk	18/54 (33)	32/49 (65)
Rice	7/43 (16)	11/40 (28)
Wheat	15/38 (39)	24/42 (57)
Codfish	4/44 (9)	11/41 (27)
Soy	15/48 (31)	18/41 (44)
Aeroallergen positive (>3-mm wheal diameter) skin prick test result, No. (%)		
Grass	6/50 (12)	10/39 (26)
<i>Dermatophagoides farinae</i>	6/58 (10)	6/42 (14)
<i>Dermatophagoides pteronyssinus</i>	7/58 (12)	8/42 (19)
Cat dander	11/54 (20)	12/42 (29)
Dog dander	6/46 (13)	10/35 (29)

Abbreviations: MiCMAC, Milan Cow's Milk Allergy Cohort; sIgE, specific IgE.

Presenting symptoms uncorrelated to duration of disease were urticaria and/or angioedema, gastrointestinal symptoms, anaphylaxis, and atopic dermatitis. Cosensitization rates observed among MiCMAC patients (Table 2) were 79.5% (89/112) with food allergens and 37.5% (42/112) with inhalant allergens. Sensitization to beef occurred in 43.3% of patients. The more prevalent cosensitizing foods were egg and peanut (evaluated by SPT) and egg and wheat (measured as specific IgE). The more prevalent inhalant allergen was cat dander. Table 3 reports the sensitization variables stratified by tolerance outcome at univariate analysis. At SPT, each 1-mm increment in wheal diameter to cow's milk reagent, fresh cow's milk, hen's

egg white and yolk, beef, soy, wheat, dog dander, and grass mix was associated with longer duration of disease. A single log increment of specific IgE to cow's milk, α -lactalbumin, β -lactoglobulin, casein, hen's egg white and yolk, beef, rice, soy, and wheat and total IgE determinations were similarly correlated. The eliciting dose at DBPCFC was a marker of longer duration of disease, with each log decrement yielding an inverse HR of 1.46. Family history of allergic disease, age at presentation, sex, and z scores for birth weight and length and weight and length for age were not significantly correlated with duration of disease. At SPT, α -lactalbumin, β -lactoglobulin, casein, peanut, codfish, rice, house dust mites, and cat dander

Table 3. Variables Significantly Associated With Longer Duration of Disease in the MiCMAC Cohort at Univariate Analysis (Skin Prick Test Wheal Diameters, sIgE Concentrations, and Oral Food Challenge Outcome)

Factor	Tolerance	Intolerance	HR (95% CI) ^a	P value
Positive bovine allergen skin prick test results, mean (SD)				
wheal diameter, mm				
Fresh cow's milk	6.43 (2.62)	8.87 (3.40)	1.21 (1.10–1.33)	<.001
Beef	3.67 (0.82)	6.00 (2.00)	1.41 (1.10–1.81)	.008
Bovine allergen sIgE concentrations, mean (SD), kUI/L				
Cow's milk	6.25 (9.88)	20.76 (27.09)	1.85 ^b (1.25–2.73)	.002
α-Lactalbumin	2.32 (3.44)	12.75 (22.31)	1.66 ^b (1.16–2.38)	.006
β-Lactoglobulin	2.04 (3.55)	8.04 (14.90)	1.81 ^b (1.23–2.67)	.003
Casein	4.64 (8.18)	14.80 (24.66)	2.05 ^b (1.37–3.07)	<.001
Beef	0.67 (1.90)	3.14 (6.91)	5.46 ^b (1.94–15.38)	.001
Positive nonmilk food allergen skin prick test results, mean (SD) wheal diameter, mm				
Egg white	7.62 (3.25)	9.16 (3.84)	1.07 (1.01–1.13)	.03
Egg yolk	6.00 (2.61)	6.47 (2.87)	1.10 (1.02–1.18)	.02
Wheat	8 (2.83)	5.33 (1.30)	1.23 (1.02–1.49)	.03
Soy	4.00 (1.41)	4.83 (1.86)	1.55 (1.11–2.17)	.009
Food allergen sIgE concentrations in the 0.35–100 range of the assay, mean (SD), kUI/L				
Egg white	6.26 (14.28)	17.99 (27.59)	2.27 ^b (1.47–3.48)	<.001
Egg yolk	1.01 (2.57)	3.16 (5.21)	3.33 ^b (1.62–6.85)	.001
Rice	0.52 (2.11)	1.21 (3.78)	6.37 ^b (1.56–26.32)	.01
Wheat	2.29 (5.82)	11.40 (24.33)	2.20 ^b (1.15–4.24)	.02
Soy	1.06 (2.65)	1.66 (3.17)	2.60 ^b (1.15–5.88)	.02
Total sIgE concentrations, mean (SD), kUI/L	149.86 (288.61)	210.17 (316.03)	2.33 ^b (1.40–386)	.001
Positive skin prick test results for aeroallergens, mean (SD) wheal diameter, mm				
Grass	4.00 (1.10)	5.00 (2.05)	1.22 (1.01–1.48)	.04
Dog dander	3.33 (0.52)	3.70 (0.82)	1.32 (1.03–1.68)	.03
No. of patients according to eliciting dose at diagnostic oral food challenge, mL				
0.1	4	10		
0.4	5	5		
1.4	14	6		
4.4	13	8		
14.4	11	9		
44.4	3	9		
144.4	9	6		
For 1-log decrement in dose			1.46 (1.04–2.03)	.03

Abbreviations: CI, confidence interval; HR, hazard ratio; MiCMAC, Milan Cow's Milk Allergy Cohort; sIgE, specific IgE.

^a Calculated with the *t* test for independence of 2 group means (2-tailed).

^b HR for log-transformed value.

allergens did not yield significant associations. Symptoms at diagnostic challenge included urticaria and/or angioedema, asthma and/or rhinitis, and immediate gastrointestinal symptoms after cow's milk ingestion, atopic dermatitis, and anaphylaxis, but no significant relationship emerged to correlate with delayed onset of tolerance.

Multivariate Analysis for Predictors of Duration of Disease

In the multivariate duration model, each 1-mm increment in wheal diameter at SPT with fresh milk (HR, 1.18; 95% CI, 1.07–1.31; *P* = .002) and the finding of a positive SPT result with soy (HR, 6.99; 95% CI, 1.56–31.25; *P* = .01) were the only 2 factors predictive of CMA persistence to reach statistical significance.

DISCUSSION

The MiCMAC Study

MiCMAC is a prospectively evaluated cohort studying the onset of tolerance in infants and children diagnosed as having immediate CMA at DBPCFC. MiCMAC demographics and patient characteristics (referrals) do not differ from other studies^{1,2,4,6,20,21} in average duration of CMA, which ranges from 12 to 30 months in the literature, although not all investigations are DBPCFC controlled.^{4,8} The mean age at diagnostic challenge in other studies is generally lower than in MiCMAC and ranges from 4.8⁴ to 7.6 months.¹ This discrepancy may reflect the specific context of Italian health care. In this country, a generalist

pediatric physician can take over the whole management of CMA in children without performing DBPCFCs.^{22–24} A longer breastfeeding period is also characteristic of the Italian infant population^{25,26} and may thus account for an older age at diagnosis.

Univariate Analysis for Independent Predictors of Tolerance

At univariate analysis, HRs were calculated for all variables and for single millimeter increments in SPT wheal diameter and for each log increment in specific and total IgE determinations and DBPCFC eliciting dose in a scalar quantification of risk of longer duration of disease. Asthma or rhinitis (as presenting symptoms), cosensitization to bovine proteins and other food and nonfood allergens, total IgE levels, and the log decrement in DBPCFC dose were significant prognostic indices for the natural history of tolerance to cow's milk in MiCMAC and connoted CMA of longer duration.

Cosensitization and Comorbidities

The finding that all patients with asthma and rhinitis also developed skin and/or gastrointestinal symptoms (data not shown) after the ingestion of food allergens shows the involvement of more than 1 target organ system. We interpret this as the other side of a clinical coin described by Illi and coworkers in 1 of the multicenter allergy studies in which children with atopic asthma with early skin and concomitant respiratory symptoms ran an increased risk (odds ratio, 2.84) of persistent asthma by the age of 7 years.²⁷ In children with CMA, in a mirror fashion, early respiratory symptoms are associated with longer duration of disease. Longer duration of CMA is similarly predicted independently by sensitization to some protein fractions on the panel of bovine allergens, although only a subset of patients underwent SPTs or specific IgE determinations with beef. The previously reported prognostic significance of casein sensitization^{28–30} is confirmed by the present study.

Among cosensitizations, egg, soy, rice, wheat, dog dander, and grass mix were similarly and incrementally linked with duration of disease. Recognition of a wide panel of ingestant and inhalant allergens, lower DBPCFC threshold dose, and total IgE concentrations appear to be the biological prerequisites that correlate with tolerance deferral in the MiCMAC study and coexpress the severity of atopy in these children.

Multivariate Analysis for Predictors of Duration of Disease

The multivariate duration model did not substantially alter the findings of univariate analysis. The independent predictive factors to emerge from this model are wheal diameter increment at SPT with fresh milk and sensitization to soy. Sensitization to less prevalent allergens is known to predict the development of asthma in birth cohorts.^{27,31} In the context

where soy is a less prevalent allergen, it is a powerful predictor of persistence of CMA.

CONCLUSION

The fate of all children with a positive DPPCFC result is not fixed. This is the first study, to our knowledge, that used 2 DBPCFC procedures as end points to identify incremental biological predictors and clinical factors that contribute to longer CMA in a duration model with referrals. In MiCMAC, the worst atopy connoted longer deferral of tolerance within observation. In this experimental protocol, the larger wheal size in the skin test with fresh cow's milk and the early acquisition of soybean sensitization suggested an atopic dyad of prognostic significance in the natural history of cow's milk tolerance. These 2 variables achieve clinical utility in planning DBPCFC-controlled tolerance schedules for children with CMA. The linkage between this defined clinical atopy and prognosis emerging from MiCMAC, together with the quantification of biological markers of longer disease, narrows the gauge for identifying candidates for intervention aimed at modifying the natural history of the disease.

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