

Original article

A protocol for oral desensitization in children with IgE-mediated cow's milk allergy

Objectives: To desensitize children with severe immunoglobulin (Ig)E-mediated cow's milk allergy in a period of 6 months by introducing increasing daily doses of cow's milk (CM) in order to enable the child to assume 200 ml of CM daily, or to induce tolerance of the highest possible CM dose.

Study design: Twenty-one children at least 6 years old with severe IgE-mediated CM allergy were admitted to the study. A convincing history of IgE-mediated CM allergy or a positive double-blind placebo-controlled food challenge with CM confirmed the diagnosis. Oral desensitization was performed with increasing doses starting from 0.06 mg of CM proteins.

Results: Overall, 15 of 21 children (71.4%) achieved the daily intake of 200 ml during a 6-month period; three of 21 children (14.3%) tolerated 40–80 ml/day of undiluted CM; three of 21 children (14.3%) failed the desensitization because they presented allergic symptoms after ingesting minimal amounts of diluted CM.

Conclusions: We successfully desensitized 15 of 21 children with severe IgE-mediated CM allergy in a period of 6 months. We stress the importance of the partial outcome in those three of 21 children who could not reach the maximum amount of 200 ml/day of whole CM, but were able to tolerate 40–80 ml/day of CM. In this way we dramatically reduced the risk of severe reactions after accidental or unnoticed introduction of low quantities of CM. We do not propose generalizing this method beyond trained staff.

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Key words: anaphylaxis; cow's milk allergy; end point skin prick test; food allergy; oral desensitization; therapy of cow's milk allergy.

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Adverse reactions to food pose continuous challenges to daily practice. Until now, the basic treatment for adverse reactions to food has been to avoid the incriminated food (1–3). However, this approach is difficult in the case of very common foods. Moreover, elimination diets can induce psychological problems, particularly in children with potential life-threatening reactions.

Due to the lack of standardized diagnostic procedures and limited knowledge of the pathogenesis causing several kinds of adverse reaction to food, many committees (4–6) have attempted to classify these reactions on the grounds of various criteria (clinical, pathogenetic, etc.). For the purposes of this study, the definitions proposed in the EAACI's position paper published in 2001, were used (6). According to these definitions, the existence of an immunoglobulin (Ig)E reaction to a certain food (*IgE-mediated food allergy*) can reasonably be supposed when symptoms are

immediate or life threatening and are accompanied by a highly positive skin prick test (SPT) and/or high radioallergosorbent test (RAST) class (2). This does not exclude IgE (co-)participation in delayed food-induced reactions, nor in nonlife-threatening food-induced diseases (7).

As the incidence of food allergy is higher in young children (8–10) than in adults (11, 12), it is clear that some children outgrow their food allergies. As reviewed by Høst (1, 13), it has been shown that infants with *non-IgE-mediated cow's milk (CM) allergy* have a higher recovery rate compared with infants with high IgE levels to CM proteins (*IgE-mediated CM allergy*). In addition, those with IgE-mediated CM allergy run a greater risk of developing other food allergies, asthma and rhinoconjunctivitis before the age of 10. In fact, 86% of children with IgE-mediated CM allergy recovered at the age of 5, but 14% of children showed persisting symptoms at the ages of 5 and 10. This is why a 6-year-old (or older) child who is still suffering from an IgE-mediated allergy to CM, has a low probability of recovering spontaneously in a short time.

Abbreviations: CM, cow's milk; DBPCFC, double blind placebo-controlled food challenge; RAST, radioallergosorbent test; REAST, reverse enzyme allergosorbent test; SPT, skin prick test.

Aims

To desensitize a group of children with severe IgE-mediated CM allergy in a period of 6 months by introducing increasing daily doses of CM.

Our targets were: (a) to enable the child to take 200 ml of CM daily, or (b) to identify the maximum amount of CM tolerated by the child, in order to minimize or eliminate severe reactions because of accidental or unnoticed introduction of low quantities of CM.

Patients, materials and methods

Twenty-one children (15 male, six female, with a median age 6 years 11 months, and actual ages ranging 5 years 3 months to 10 years 2 months) with severe IgE-mediated CM allergy were admitted to the study from March 1999 to December 2000.

The children admitted had to be at least 6 years old in order to be reasonably sure that oral tolerance to CM was not naturally achieved (13). Because of parental pressure, four children (PetM, CF, CS, and DV) were admitted although they were slightly younger than six (Table 1).

A detailed clinical history was obtained, recording: sex, age, family and personal history of allergies, symptoms after previous ingestion of CM.

Skin prick tests with commercial common inhalant allergens (*Dermatophagoides pteronyssinus*, *Lolium perenne*, *Parietaria officinalis*, *Olea europea*, *Alternaria tenuis*, cat dander), with commercial food allergens (casein, β -lactalbumin, egg white, soy, wheat, and cod) (Lofarma, Milano, Italy) and with the same hydrolysate formula used for the double blind placebo-controlled food challenge (DBPCFC) (placebo), were done. The SPTs were performed on the volar aspect of the forearms with a plastic lancet with a 1 mm point, pricking through, for 1 s, a drop of the allergen extract, which was wiped away immediately afterwards with a gauze. Glycerine with histamine 1 : 1.000 was used as a positive control and glycerine alone as a negative control. SPTs were read at 15 min and wheals measuring 3 mm or more in diameter and at least 3 mm more than the negative control, were considered positive.

End point SPTs with casein and β -lactalbumin extracts were performed. On a different day of standard SPTs, at the beginning of the desensitization protocol with CM, and at the end of the protocol (after a 20-day interruption of cetirizine, see below), two series of SPTs with scalar amounts of casein and β -lactalbumin extracts were performed, starting with 1 : 20 w/v of casein and β -lactalbumin (Lofarma). Casein SPTs were performed on the right forearm, and β -lactalbumin on the left, applying one drop of casein and β -lactalbumin extracts diluted 1 : 1, 1 : 10, 1 : 100, 1 : 1 000 and 1 : 10 000. As for standard SPTs, these reactions were read at 15 min. Results were calculated as the mean of the orthogonal diameters of the wheal.

Double blind placebo-controlled food challenge tests

The DBPCFCs were performed in hospital as proposed by Niggemann et al. (14), with some modifications. The verum meal consisted of 125 ml of whole CM + 125 ml of casein (or whey) hydrolysate + 1 g of banana flavoring (total 250 ml). The placebo meal consisted of 250 ml of casein (or whey) hydrolysate + 1 g of banana flavoring (total 250 ml). A specific brand of casein (or whey) hydrolysate was not chosen. Instead, the one previously tolerated by the child, was used.

The children were in the fasting state and had been withdrawn from systemic drugs at least 7 days previously. In 2 days, according to a random, computerized schedule, they were given the meal containing CM (or placebo) by means of a syringe or a feeding bottle covered with metal foil and the reactions were always recorded by the same physician. Initially, three drops of the meal containing whole CM (or the placebo meal) were put in the lower lip fornix. Further oral doses of 0.5, 2, 5, 20, 60 and 162.5 ml were given every 15 min. When symptoms arose, the test was terminated. After 4 h observation following the last administration of the CM (or the placebo) the children were discharged.

The CM desensitization protocol

The children with a convincing history of IgE-mediated CM allergy and the children with IgE-mediated CM allergy confirmed by positive DBPCFC, were admitted to the oral desensitization trial that started after 2–3 months after the last convincing reaction to CM, or after on average 23 days after the positive DBPCFC (range 18–32 days).

The schedule consisted of administering increasing amounts of CM starting from one drop of whole CM diluted 1 : 25 with water corresponding to approx. 0.06 mg of CM proteins (Table 2). The CM doses were doubled every 7 days until day 70. Subsequently, the CM doses were doubled every 16 days in order to achieve a total daily intake of 200 ml in about 6 months. Dose doubling was carried out gradually with increasing amounts of CM between one dose and the next. When an inter-current illness intervened (common cold, viral diarrhea, fever, etc.) during the oral desensitization, the daily dose of CM was not increased and the previous dose was repeated.

The following doses were administered at the hospital: 1, 2 and 4 drops of diluted CM; 1, 2, 4 drops of undiluted CM and 1, 2, and 4 ml of undiluted CM. Parents administered the other CM doses at home. During the 6-month duration of the protocol, all the children were given 0.25 mg/kg/day of cetirizine *per os*. This drug was suspended 2 weeks after the end of the protocol (whilst children were continuing to take CM), when the child reached 200 ml/day or the maximum tolerated dose of whole CM in order to see whether symptoms occurred without the cover of cetirizine.

All the children received monthly check-ups for at least 3 months after the end of the protocol.

Blood samples

Three peripheral venous blood samples were collected: the first at the beginning of the protocol; the second after 3 months or when the child had reached a 5 ml dose of whole CM; and the third at the end of the protocol. The serum samples were stored at -20°C . For each serum sample total IgE and specific IgE for whole CM, β -lactalbumin and casein have been determined with reverse enzyme allergosorbent test (REAST), a 'capture' method consisting briefly in the binding of IgE by a specific anti-IgE antibody, the reaction with anti-IgE-biotin and allergen-biotin respectively, subsequent reaction with streptavidin-peroxidase and chromogen-substrate (15). The optical density values were read on a IgE standard curve referred to the WHO IgE international reference preparation, and expressed as kU/l. Both total and specific IgE determinations have been performed with commercial reagents (Realtest[®]-Total-IgE and Realtest[®]-Specific IgE; Lofarma). Moreover sideraemia, and hemochrome, were determined.

Feces samples

At the same time as the blood samples, hidden blood in the feces was also sought (Hema Screen, Immunostics, Inc. Ocean, NJ).

Table 1. Symptoms after double blind placebo-controlled food challenge (DBPCFC) with cow's milk (CM), after accidental ingestion of CM, and during CM desensitization

Patient	Age at the start of CM desensitization (Years, months)	Time between DBPCFC (or accidental ingestion of CM) and onset of symptoms (min)	Symptoms after DBPCFC with CM	Symptoms after accidental ingestion of CM (DBPCFC not done for 'convincing history')	Outcome of CM desensitization	Symptoms during CM desensitization (see text for more details)	Dose of CM when symptom(s) intervened
1 ChF	7, 3/12	120	Perioral angioedema, urticaria, asthma	—	*	—	—
2 DAA	6, 1/12	20	Urticaria, angioedema	—	*	—	—
3 DGP	6, 5/12	10	Erythema, urticaria	—	*	—	—
4 GF	8, 1/12	20	Pruritus, edema, urticaria, conjunctivitis, abdominal pain	—	*	—	—
5 IA	8, 6/12	160	Urticaria, abdominal pain	—	*	—	—
6 LPD	10, 4/12	15	Vomiting, abdominal pain	—	*	—	—
7 PerM	7, 10/12	90	Pruritus, erythema, perioral urticaria, vomiting	—	*	—	—
8 PetM	5, 6/12	1	Pruritus, urticaria, edema	—	*	—	—
9 CF	5, 3/12	30	—	Urticaria, perioral angioedema, severe asthma	†	Temporary nasal obstruction	From 7 to 12 ml of undiluted CM
10 CS	5, 6/12	60	—	Acute diarrhea, vomiting, profuse sweating, soles pruritus	†	Rhinitis, abdominal pain	From 45 to 55 ml of undiluted CM
11 ML	10, 2/12	15–20	Pruritus, urticaria, angioedema, cough	—	†	Abdominal pain	From 1 to 5 ml of undiluted CM
12 PI	7, 3/12	15	Urticaria	—	†	Temporary throat pruritus	From 10 drops of CM diluted 1 : 25 to two drops of undiluted CM
13 MR	10	50	Severe asthma	—	†	Temporary throat pruritus	From two drops to 2 ml of undiluted CM
14 RR	8, 7/12	25	Pruritus of tongue, edema	—	†	Temporary erythema (hands and popliteal and elbow folds)	From 9 to 15 ml of undiluted CM
15 TS	6	1	Erythema, urticaria	—	†	Temporary soles pruritus	From 80 to 85 ml of undiluted CM
16 DOA	6, 7/12	120	Erythema, edema, rhinitis, vomiting, rhinitis (1)	—	‡	Moderate asthma	47 ml of undiluted CM
17 PM	7, 6/12	70	Erythema, cough (2)	—	‡	Angioedema	45 ml of undiluted CM
18 VS	8, 3/12	1	Tickly throat, cough, asthma (3)	—	‡	Asthma, whole body erythema	90 ml of undiluted CM
19 DV	5, 10/12	10	Rhinitis, pharyngeal pruritus	—	§	Urticaria, throat pruritus	10 drops of undiluted CM
20 GS	6, 11/12	1	Vomiting	—	§	Throat pruritus, epigastric pain, vomiting	2 drops of diluted CM
21 PD	6, 1/12	70	—	Severe asthma	§	Moderate asthma	18 drops of undiluted CM

Outcome of CM desensitization:

* Tolerates 200 ml of whole CM per day.

† Tolerates 200 ml of whole CM per day (presented some symptoms which occurred in a short time after ingestion of CM and persisted for <2 h).

‡ Tolerates between 40 and 80 ml of whole CM per day. During the DBPCFC they had symptoms: (1) with 60 ml of verum (= 30 ml of CM); (2) with 5 ml of verum (= 2.5 ml of CM); (3) with 30 ml of verum (= 15 ml of CM).

§ Oral CM desensitisation failed (immediate onset of allergic symptoms after a few doses).

Table 2. Schedule of oral cow's milk (CM) desensitization

Day of oral desensitization	Doubling of CM doses	Drops of CM (diluted 1 : 25)	Drops of CM (not diluted)	Milliliters of CM (not diluted)	CM proteins amount (g)
1	—	1			0.00006
7	1	2			0.00013
14	2	4			0.00026
21	3	8			0.0005
28	4	16			0.001
35	5	32			0.002
42	6	64			0.004
49	7	—	5		0.008
56	8	—	10		0.016
63	9	—	20		0.03
70	10	—	—	2	0.06
86	11	—	—	4	0.13
102	12	—	—	8	0.26
118	13	—	—	16	0.5
134	14	—	—	32	1.0
150	15	—	—	64	2.0
166	16	—	—	128	4.1
180	(17)	—	—	200	6.4

Every dose doubling was reached gradually from the previous to the next dose.

Statistical analysis

The statistical analysis was performed using the Wilcoxon matched-pairs signed-ranks test.

Results

Sixteen of the 21 children (76%) had a positive family history for atopic diseases. Fifteen of 21 children have had atopic dermatitis in the past, eight of 21 children suffered from conjunctivitis, 17 of 21 children from allergic rhinitis and 15 of 21 children from asthma. All the included children have had symptoms after previous ingestion of CM (pruritus, erythema, urticaria, angioedema, perioral dermatitis, atopic dermatitis, vomiting, abdominal pains, diarrhea, conjunctivitis, rhinitis, asthma and anaphylaxis).

Skin prick tests with common allergens

All the children underwent SPTs with commercial common inhalant allergens presenting positivities to *D. pteronyssinus* (61.1%), *L. perenne* (71.4%), *P. officinalis* (47.6%), *O. europea* (42.8%), *Alternaria alternata* (19%), cat dander (23.8%), casein (90.5%), lactoalbumin (95.2%), egg white (76.2%), and cod (23.8%). No child was positive to wheat or soy.

Skin prick tests with casein (or whey) hydrolysates

All the children underwent SPT with the casein (or whey) hydrolysate formula previously tolerated by them, and all resulted negative.

Table 3. Outcome of the end point skin prick tests (SPTs) with casein and β -lactoalbumin at the beginning (pre), and at the end (post) of the desensitization period. The cutaneous sensitivity for both casein and β -lactoalbumin significantly decreased after the 6-month period of desensitization ($P < 0.001$, matched-pairs Wilcoxon test). The figures express the sum of the reactions to the various dilutions

	End point SPTs (mm) (Casein)			End point SPTs (mm) (β -lactoalbumin)		
	Pre	Post	<i>P</i> -value	Pre	Post	<i>P</i> -value
Average ($\pm$ SD)	29 ($\pm$ 14)	15 ($\pm$ 6)	<0.001	31 ($\pm$ 10)	16 ($\pm$ 7)	<0.001
Median	26	15		32	15	
Range	18–50	5–25		17–49	5–29	

End point SPTs with casein and β -lactoalbumin

Eighteen of the 21 children were submitted to End Point SPTs with casein and β -lactoalbumin extracts. The cutaneous sensitivity for both casein and β -lactoalbumin significantly decreased after the 6-month period of desensitization ($P < 0.001$, matched-pairs Wilcoxon test) (Table 3) in the children who completed the protocol.

DBPCFC

Three of the 21 children did not undergo the DBPCFC because of a convincing history of severe reaction after the ingestion of minimal amounts of CM (Table 1). In particular, CF presented urticaria, perioral angioedema and asthma after ingestion of *mozzarella* (a soft curd cheese), cooked ham or bread (both containing hidden milk), 2 months before starting oral desensitization; CS

Table 4. Results of dosage of total and specific IgE [reverse enzyme allergosorbent test (REAST)] for whole cow's milk (CM), casein and β -lactalbumin before (pre) and after (post) the oral desensitization. All the differences were not significant (N.S.)

	REAST (kU/l) – total IgE			REAST (kU/l) – specific IgE (whole CM)			REAST (kU/l) – specific IgE (β -lactalbumin)			REAST (kU/l) – specific IgE (casein)		
	Pre	Post	P-value	Pre	Post	P-value	Pre	Post	P-value	Pre	Post	P-value
Average ($\pm$ SD)	1043 ($\pm$ 942)	1291 ($\pm$ 1371)	N.S.	17.1 ($\pm$ 29.7)	12.7 ($\pm$ 18.6)	N.S.	19.4 ($\pm$ 41.3)	14.8 ($\pm$ 27.6)	N.S.	1.8 ($\pm$ 2.7)	1.5 ($\pm$ 2.6)	N.S.
Median	808	804		3.9	3.3		4.1	3.2		0.9	0.6	
Range	66–3233	90–5251		0.01–94.8	0.24–52.4		0–147	0–85		0–10.4	0–10.92	

suffered from diarrhea, vomiting and profuse sweating and soles pruritus an hour after ice-cream ingestion or unnoticed CM ingestion due to CM contaminated cutlery, 2.5 months before starting oral desensitization; PD suffered from severe asthma after accidental inhalation and/or ingestion of CM, 3 months before starting oral desensitization.

Eighteen of the 21 children underwent the DBPCFC showing with the verum meal one or more of the following symptoms: pruritus, urticaria/angioedema, erythema, tickly throat, pharyngeal pruritus, vomiting, abdominal pain, conjunctivitis, rhinitis, coughing, and asthma (Table 1). All the DBPCFC with placebo resulted negative.

The desensitization protocol

Overall, 15 of the 21 children (71.4%) achieved the daily intake of 200 ml. Indeed, eight of 15 children reached the full CM dose without showing any symptom. The other seven of 15 children presented some symptoms (Table 1) which started shortly after ingesting CM and persisted for < 2 h.

No additional drug was added to cetirizine to control these symptoms. All these children did not present any symptoms 2 months after stopping cetirizine and continued taking whole CM.

Three of 21 children (14.3%) were able to tolerate 40–80 ml/day of undiluted CM. Increasing the intake of CM to 47 ml provoked asthma after about 10 min in patient DOA. However he then tolerated 40 ml/day of whole CM without symptoms. Patient PM suffered from urticaria and perioral angioedema about 3 min after he had taken 45 ml of whole CM. He then tolerated 40 ml/day of whole CM. Twenty minutes after taking 90 ml of whole CM, patient VS suffered from asthma and whole body erythema. He then took 80 ml of whole CM without any symptoms. These three children did not present any symptoms 2 months after stopping cetirizine although they were continuing to take their daily dose of whole CM.

Three of the 21 children (14.3%) could not follow the desensitization schedule because they presented: rhinitis and tickly throat with 10 drops of undiluted CM (patient DV; we tried twice before stopping

desensitization); throat pruritus, epigastric pain, vomiting with two drops of CM diluted 1 : 25 (patient GS; we tried three times before stopping desensitization); moderate asthma about 30 min after taking 18 drops of undiluted CM (patient PD; we tried twice before stopping desensitization).

As a result of inter-current illnesses, the actual duration of the desensitization protocol was longer than expected (180 days) with a mean of 201 days and a range of 183–234 days.

Specific IgE for whole CM, casein and β -lactalbumin (REAST)

The differences between the values of REAST for total IgE and specific IgE for whole CM, casein and β -lactalbumin at the beginning of the protocol, after 3 months and at the end of the protocol were not significant ($P > 0.1$, matched-pairs Wilcoxon test) (Table 4).

No significant differences in hemocrome and sideraemia values were observed between the beginning and the end of the protocol.

There was no trace of hidden blood in the feces of any of the children.

Discussion

Once CM allergy is diagnosed, a strict elimination diet is, at present, the only proven therapy (2). This approach is seemingly simple. However, if the food allergy is persistent and the elimination diet has to be prescribed for a long period of time, malnutrition, eating disorders, and psychological problems can arise (2, 16). Moreover, the elimination of CM from the diet is sometimes difficult because it can be hidden and not necessarily labeled in 'non-dairy' commercial foods (2, 17). On the contrary, a strict elimination diet is mandatory if low quantities of CM can be life-threatening (18, 19). For these reasons, alternative therapies to elimination diets have been experimented. The possibility of obtaining food tolerance through oral desensitization has been attempted for several foods and isolated, and successful experiences using increasing oral doses of CM have already been reported as reviewed by Patriarca et al. (20).

In his own work, Patriarca et al. (20, 21) began with four drops of diluted CM (corresponding to about 0.3 mg of CM proteins). The CM doses were then increased until the total daily dose of 120 ml of whole CM was reached on day 104. At the beginning of the treatment, oral sodium cromoglycate was administered 20 min before CM ingestion. If no reaction occurred, this pretreatment was withdrawn within a few days. Oral CM desensitization was successful in four of six children. The remaining two of six children dropped out because of lack of compliance. During the treatment five of six patients experienced urticaria, there was one case of angio-oedema and one case of asthma with abdominal pain. These symptoms were controlled by oral antihistamines.

Bauer et al. (22) reported an oral rush desensitization to CM in a 12-year-old girl who suffered from generalized urticaria and Quincke's edema after CM ingestion. They started with 1 ml of CM diluted 1 : 100 (corresponding to about 0.32 mg of CM proteins). By doubling doses, they successfully reached the dose 200 ml of whole CM at day 5. The Authors suggested performing their 5-day schedule under strict medical control.

We tried to desensitize 21 children with severe IgE-mediated CM allergy within a period of 6 months, by introducing increasing daily doses of whole CM. With the exception of the very first doses, our schedule foresaw CM introduction at home. Because of the length of our protocol, we decided to perform the study in a open way. In order to be reasonably sure that spontaneous oral CM tolerance would not be reached in the immediate future, we chose an entry age of about six for admittance to oral desensitization. Indeed, the prognosis of CM allergy is quite promising, with a mean recovery rate of over 80% at 3 years as shown in some prospective observational studies (9, 23–25). On the contrary, some Authors (26) have recorded the persistence of chronic diarrhea because of CM allergy at the age of six in 12 of 37 children (32.5%) with RAST and SPTs highly positive for CM (*IgE-mediated CM allergy*). Because of differences in study design, diagnostic criteria and entry age in other reports, the recovery rates varies between 33% (27) and 98% (28). Generally, the higher the age at inclusion in the study the lesser the probability of becoming tolerant to CM. In addition children whose food sensitivity is diagnosed at a later ages, tend not to outgrow the problem (29).

Desensitization was completely successful in 15 of 21 children (71.4%). Eight of these children achieved the full CM intake without showing any symptoms. The other seven children presented some symptoms (Table 1) shortly after ingestion of CM which persisted for <2 h. Because of the open protocol, we cannot be sure that those symptoms was caused by CM introduction. In any event, they were temporary, disappeared in a short time and no additional drugs (other than cetirizine) were required to control them. Furthermore, they did not interfere with the desensitization protocol and did not

appear again after 2 months when cetirizine was suspended.

As reported by Yunginger et al. (18) the majority of anaphylactic reactions occur outside the home where one is less certain of the ingredients of food consumed. In such cases, it is likely that the amount of hidden food required to cause a reaction is very low. Moreover, the still unresolved issue of food labeling must be taken into account (18, 43). We therefore favorably view the results of the three of 21 children (14.3%) in our protocol who could not reach the final dose of 200 ml/day of CM but were able to tolerate 40–80 ml/day of undiluted CM. It could be speculated that in these highly sensitive children even low doses of CM could induce local inflammation of the gastrointestinal tract with the risk of inducing new sensitizations, but we did not observe any clinical reaction even 2 months after we had reached the maximum tolerated dose and after cetirizine had been suspended. In our opinion these developments show considerable progress since the children have become protected from the inadvertent introduction of low quantities of CM and, as a result, are at lower risk of severe anaphylactic reactions.

We tried to look for some difference between subjects with good, poor or no tolerance. As reviewed by Høst (1), it has been demonstrated that infants with high IgE levels to CM have a lower rate of recovery than infants with non-IgE-mediated reactions. In our study we used the REAST method instead of the RAST one. In a previous study, the correlation between REAST and RAST with inhalant allergens resulted to be satisfactory, being 0.91 for *Dermatophagoides*, 0.95 for *Parietaria*, 0.94 for grass, and 0.76 for birch (30). On the contrary, food allergens can cause some discordance problems, as the presence of allergen-specific IgG antibodies can interfere with RAST, causing false negative results, while it has no effects on REAST, as demonstrated in a REAST vs RAST comparative study on CM-specific IgE antibodies in children (31). Sampson and Ho (32) showed that the higher the RAST class of a certain food, the higher the possibility of having symptoms after introducing that food. These observations can account for the fact that the three of 21 children who could not carry out the oral desensitization as they had severe symptoms from the first diluted CM administrations, had a REAST class 4 or 5 for casein and/or β -lactalbumin. Similarly, the three of 21 children who could tolerate a CM dose of 40–80 ml/day had a REAST class ranging between 3 and 5. Conversely, one child (PetM) with class REAST 5 was able to tolerate 200 ml of whole, undiluted CM at the end of the protocol. All the children (10 of 21, 42%) with REAST class 1 or 2 were able to complete the desensitization protocol. We therefore conclude that the higher the REAST class for CM, the higher the possibility of failure in the desensitizing protocol. Nevertheless, according to our experience, exceptions to this rule have to be taken into account and the outcome of the desensitization

protocol cannot be predicted on the grounds of REAST values alone.

Oral food tolerance is induced by several mechanisms including clonal deletion, clonal anergy, and active cellular suppression (33). It seems the one of the determining factors is the dose of fed antigen (33–38). Low doses of antigen mostly induce active suppression, whereas high doses favor deletion and anergy. Active suppression is mediated both by the induction of regulatory T-cells in the GALT and the local secretion of suppressive cytokines such as transforming growth factor β , interleukin (IL)-4 and IL-10 following the antigen-specific trigger (33, 39–41). Referring to other works for in-depth knowledge of the oral food tolerance (42), we can assume that in the children in our study who were able to take the full or partial CM dosage, some of the mechanisms regulating oral tolerance are involved.

We did not investigate gastrointestinal mediators, but demonstrated a significant reduction of cutaneous sensitivity to CM by means of the End Point SPTs with scalar amounts of casein and β -lactalbumin before and after the desensitization period (Table 3). We were not able to demonstrate a similar reduction in REAST values (Table 4).

It is worth noting that our children tolerated CM even 2 months after stopping cetirizine and continuing to take CM. We do not know whether they really could tolerate CM without previous treatment with an antihistamine, but it was given in order to render the protocol safer as it was performed at home. Otherwise, we looked at the final result, that is, to desensitize the child.

Conclusions

Adverse reactions to foods are a continuous challenge and until now the basic treatment has been to avoid the incriminated food. We successfully desensitized 15 of 21 children aged over 6 years and suffering from severe IgE-mediated CM allergy in a period of about 6 months, by introducing increasing daily doses of CM. Our protocol failed with three of 21 children who had the highest REAST class for CM, although one child with REAST class 5 was able to tolerate 200 ml of whole undiluted CM by the end of the protocol.

We stress the importance of the partial outcome in those three of 21 children, who, could not reach the maximum amount of 200 ml/day of whole CM, but were able, nevertheless, to tolerate 40–80 ml/day of CM. In this way we dramatically reduced the risk of severe reactions after accidental or unnoticed introduction of low quantities of CM.

This protocol is time consuming, but offers the advantage that it can be performed at home. Although we believe this methodology is quite safe, we do not propose generalizing it beyond trained staff, and maintain that standardization of these methodologies is necessary.

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