



Allergologia et immunopathologia

Sociedad Española de Inmunología Clínica,
Alergología y Asma Pediátrica

www.all-imm.com



ORIGINAL ARTICLE

OPEN ACCESS

Outcome of food intake and nutritional status after discontinuation of a cow's-milk-free diet post negative oral food challenge in infants and children

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Received 23 June 2021; Accepted 30 September 2021

Available online 1 January 2022

KEYWORDS

body height;
children;
infant development;
milk hypersensitivity;
nutritional status;
food intake

Abstract

Objective: To evaluate the outcome of food intake and nutritional status post discontinuation of a cow's-milk-free diet after a negative oral food challenge.

Methods: This was a prospective, uncontrolled study that evaluated food intake and nutritional status of a cohort of 80 infants and children under 5 years of age. Food intake and nutritional status were evaluated on the day of the oral food challenge test and after 30 days. Weight and height were measured on the day of the test and after 30 days.

Results: The mean age of the patients was 18.7 ± 12.4 months, and 58.7% were male. After discontinuation of the elimination diet, the children showed daily intake increases in ($P < 0.001$), protein ($P < 0.001$), carbohydrates ($P = 0.042$), calcium from foods ($P < 0.001$), calcium from foods and supplements ($P < 0.001$), phosphorus ($P < 0.001$), and vitamin D from foods ($P = 0.006$). The Z-scores ($n = 76$) on the day of the oral food challenge test and 30 days after restarting the consumption of cow's milk were as follows: weight-for-age ($P < 0.001$) and height-for-age ($P < 0.001$), respectively.

Conclusion: Cow's milk protein in the diet was associated with increased intake of energy, proteins, carbohydrates, calcium, phosphorus, and vitamin D, in addition to an increase in the Z-scores for weight-for-age and height-for-age.

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<https://doi.org/10.15586/aei.v50i1.471>

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Introduction

Cow's milk allergy (CMA) is the most common food allergy in infants,¹ with an estimated prevalence in developed countries ranging from 0.5% to 3.0% in the first years of life.^{2,3} In Brazil, an epidemiological study in pediatric gastroenterologist offices revealed that the incidence and prevalence of CMA are 2.2% and 5.4%, respectively.⁴

To confirm diagnosis of CMA and the development of oral tolerance to cow's milk, a double-blind placebo-controlled food challenge is considered to be the gold standard.^{3,5-8} However, this method is difficult to perform, especially when a reaction is delayed, that is, an allergy can reappear up to 4 weeks from the beginning of re-exposure to the allergen.^{1,3,5,6-8} Its application may have greater value in situations in which clinical manifestations are immediate and subjective, and patients may be susceptible to food intake and thus may have sensations not linked to re-exposure to the allergen.^{8,9} In this context, especially in children up to 3 years of age, open food challenge (OFC) is recommended as the first option in clinical practice,^{1,6,8,10} enabling a significant number of patients to be fed a cow's milk protein elimination diet to discontinue the diet.^{11,12}

The allergenic protein elimination diet is the treatment for almost all infants and children with CMA. However, the cow's milk elimination diet when improperly used can impact and compromise nutritional status.^{1,3,6,8} Taking into account the higher cost of special formulas for infants with CMA and the difficulties in feeding a diet without cow's milk protein, the ideal situation is to use such a diet for the strictly necessary period. Thus, the performance of the OFC is very important to prove the diagnosis of CMA and to characterize the development of oral tolerance in patients given an elimination diet. It should be highlighted that an exclusion diet is associated with a lower quality of life.^{7,8}

Studies conducted in Brazil and other countries have shown that energy and nutrient intake, especially calcium, are lower in children with CMA than in children without food allergies.¹³⁻²¹ However, there is little information on the impact of interrupting an elimination diet on food intake and nutritional status. In the literature, only one retrospective study²², conducted in Japan, evaluated the growth of 58 children after the discontinuation of a cow's-milk-free diet, which found an increase in height-for-age Z-score. In that study, however, food intake was not evaluated. The authors hypothesized that lower calcium intake could be related to lower values of height-for-age Z-score. To our knowledge, no study has evaluated the changes in food intake provided by the discontinuation of the cow's milk elimination diet.

Therefore, the objective of this study was to evaluate the outcome of food intake and nutritional status after discontinuation of a cow's-milk-free diet post a negative OFC.

Materials and Methods

Study design

This was a prospective, uncontrolled study that evaluated food intake and nutritional status of a cohort of infants and

children during a cow's milk protein elimination diet and on the 30th day after the discontinuation of such a diet post a negative OFC.

The study was approved by the Research Ethics Committee of UNIFESP under number 0718/10. All the guardians of the children participating in the study signed an informed consent form.

Population sample

All 130 children under the age of 5 years who were on a cow's milk and dairy elimination diet who underwent the OFC for cow's milk at the São Paulo Hospital of the Federal University of São Paulo, Paulista School of Medicine, between January 2015 and March 2019 were considered for inclusion in the study.

Of these 130 children, 29 (22.3%) were excluded for having a positive challenge test (6 with immediate reactions and 23 with delayed reactions).

Of the 101 that had an oral challenge test negative for cow's milk proteins, 80 (79.2%) were reassessed after 30 days.

Open food challenge

The OFC was performed in the morning, with patients fasting, under the supervision of a pediatric gastroenterologist and nutritionist. Before the test, information on the purpose of the test (diagnostic confirmation or assessment of the development of oral tolerance) was provided, and the duration of the cow's milk and dairy elimination diet, type of special substitute formula that had been used, and clinical manifestations that motivated previous possible diagnosis of allergy to cow's milk protein were obtained.

A complete physical examination was performed, and weight and height were measured before the start of the OFC. Next, 1 mL of cow's milk was administered directly into the oral cavity, avoiding contact with the labial region. After 20 min, if no adverse reaction had occurred, an additional 10 mL was administered orally. If the patient remained asymptomatic, new volumes of milk were administered twice orally in increasing volumes every 20 min (20 mL, 40 mL, and 80 mL) until the total volume of milk ingested reached 151 mL.^{8,11}

If the patient remained asymptomatic for another hour (total of 2 h and 30 min after the beginning of oral exposure to cow's milk), a new physical examination was performed. If there were no abnormalities, the patient was discharged for outpatient follow-up. After discharge, a diet with cow's milk and dairy products was followed, replacing cow's milk protein-free formula by infant formula with cow's milk protein or cow's milk. For children with acceptance difficulties, it was recommended that the infant formula or cow's milk be mixed with the formula that had been previously used, ensuring a minimum intake of 150 mL of cow's milk protein preparation per day.

Patients were instructed to call if there were any symptoms. When necessary, the patient returned for an in-person medical evaluation. If there was no reaction, the

patient returned for clinical and anthropometric evaluations and to perform a new food survey 30 days after the start of the challenge test.

If the child remained asymptomatic at the end of 30 days, that is, without signs or symptoms consistent with CMA, the OFC was considered negative.^{1,2,3}

When the patient presented clinical manifestations resulting from the ingestion of cow's milk protein, the test was considered positive.^{1,3,6,7,8} Reactions that occurred up to 2 h after the beginning of re-exposure to cow's milk were considered immediate. The reactions that occurred later, until the 30th day, were considered delayed.^{1,24}

Assessment of nutritional status

Weight and height were measured and classified based on standards provided by the World Health Organization,²⁵ on the day of the OFC and on outpatient follow-up after 30 days. The classification of nutritional status was based on anthropometric indices, weight-for-age (W/A), weight-for-height (W/H), height-for-age (H/A), and body mass index-for-age (BMI/A), with the aid of Anthro software, version 3.2.2. Weight and height measurements on the 30th day were performed without knowledge of the measurements obtained on the day of the OFC.

Food intake

Food intake was assessed by means of a 24-h recall survey on the day of the OFC and 30 days after lifting the restriction of cow's milk in the diet. The 24-h recall was performed by a single nutritionist in both evaluations in accordance with the recommendations.^{26,27} To help quantify the size of the portions, a photographic album of food portions was used. If the children were taking dietary supplements, parents were asked to supply the basic supplement information, including the brand name, manufacturer, and daily dosage, to estimate nutrient intake through the dietary supplements. For the calculation of energy, protein, lipids, carbohydrates, calcium, phosphorus, iron, and vitamins A, C and D, the software Avanutri online²⁸ (Rio de Janeiro, Brazil) was used. The food intake data obtained in the two evaluations were interpreted based on dietary reference intakes (DRIs).^{29,30}

Statistical analysis

Statistical analysis was performed using SigmaStat software version 3.5 (SPSS Inc., Chicago, USA).³¹ Numerical variables were presented as absolute number and percentage. All results of statistical analyses are expressed as mean and standard deviation or as median and 25th and 75th percentiles. The paired Student's t test or the non-parametric Wilcoxon test were used for intragroup comparisons (at OFC and 30 days after the test). The McNemar test was used to verify whether there was a change in the adequacy or inadequacy ratio in food intake in relation to DRIs. The tests used are presented together with the results. The level for rejection of the null hypothesis was set at <0.05 or 5%.

Results

Demographic and clinical characteristics

Table 1 shows the clinical and demographic characteristics of the 80 children who had a negative OFC for cow's milk and who were reassessed after 30 days. Before the challenge test, most patients received an extensively hydrolyzed formula or amino acid formula. The duration of the cow's milk and dairy elimination diet was greater than 24 weeks for most patients.

Regarding the clinical manifestations that motivated the diagnostic hypothesis of CMA, gastrointestinal manifestations predominated in 95.0% (76/80), and cutaneous manifestations predominated in 13.7% (11/80) of the patients. Respiratory manifestations occurred less frequently, that is, in 10.0% (8/80). In 15% (12/80) of the children, two or more symptoms or signs were observed, and in 85% (68/80), three or more symptoms or signs were observed.

Food intake

Table 2 shows the intake of energy and macro- and micro-nutrients on the day of the OFC and 30 days after consuming a diet without any restrictions. There was a statistically significant increase in energy, protein, and carbohydrate intake. There was no variation in lipid intake. Regarding micronutrient intake, there was an increase in calcium, phosphorus, and vitamin D via food. The intake of iron, vitamin A, and vitamin D in food did not vary 30 days after

Table 1 Demographic and clinical characteristics of children with a negative oral food challenge for cow's milk.

| Variable | n = 80 | % |
|--|--------|------|
| Gender | | |
| Males | 47 | 58.7 |
| Female | 33 | 41.3 |
| Age (months) | | |
| 0-6 | 4 | 5.0 |
| 7-12 | 33 | 41.3 |
| 13-24 | 27 | 33.7 |
| 25-60 | 16 | 20.0 |
| Indication oral food challenge | | |
| Diagnostic | 16 | 20.0 |
| Tolerance | 64 | 80.0 |
| Duration of cow's-milk-free diet (weeks) | | |
| ≤12 | 5 | 6.3 |
| 13-24 | 29 | 36.2 |
| 25-47 | 26 | 32.5 |
| ≥48 | 20 | 25.0 |
| Type of cow's milk substitute | | |
| Amino acid-based formula | 31 | 38.7 |
| Extensively hydrolyzed formula | 32 | 40.0 |
| Soy protein-based formula | 8 | 10.0 |
| Soy extract-based beverage/juice | 6 | 7.5 |
| Rice drink | 1 | 1.3 |
| No substitute | 2 | 2.5 |

Table 2 Energy, macronutrient, and micronutrient intake on the day of oral food challenge and after 30 days.

| Variable | On the day | After 30 days | P |
|--|----------------------|----------------------|---------------------|
| Total energy (kcal/day) | 896.7 ± 246.9 | 961.9 ± 227.1 | <0.001 ¹ |
| Protein (g/day) | 35.5 ± 11.2 | 41.1 ± 13.9 | <0.001 ¹ |
| Lipids (g/day) | 28.7 ± 12.0 | 29.7 ± 10.3 | 0.320 ¹ |
| Carbohydrate (g/day) | 125.7 (103.1;147.5) | 130.3 (109.1; 155.7) | 0.042 ² |
| Calcium from foods (mg/day) | 495.2 (332.7; 587.2) | 712.6 (513.5; 928.3) | <0.001 ² |
| Calcium from foods and supplements (mg/day) ^a | 516.2 (351.8;608.5) | 722.9 (557.3;940.2) | <0.001 ² |
| Phosphorus (mg/day) | 463.1 (357.1;557.7) | 680.5 (489.1;786.9) | <0.001 ² |
| Iron from foods (mg/day) | 10.9 (7.7; 12.6) | 9.3 (5.9; 11.8) | 0.476 ² |
| Iron from foods + supplements (mg/day) ^b | 13.5 (10.2; 22.9) | 12.7 (9.1; 21.5) | 0.392 ² |
| Vitamin A from foods (µg/day) | 823.4 ± 505.4 | 895.0 ± 584.5 | 0.344 ¹ |
| Vitamin A from foods + supplements (µg/day) ^c | 1217.1 ± 613.4 | 1287.0 ± 715.4 | 0.360 ¹ |
| Vitamin C from foods(mg/day) | 103.2 (72.0; 151.5) | 136.4 (71.5; 178.1) | 0.100 ² |
| Vitamin C from foods + supplements (mg/day) ^d | 109.9 (76.5; 168.6) | 139.9 (81.8; 186.0) | 0.104 ² |
| Vitamin D from foods (mg/day) | 7.6 (4.1; 10.8) | 8.8 (5.0; 11.8) | 0.006 ² |
| Vitamin D from foods + supplements (mg/day) ^e | 15.6 (7.2; 21.1) | 15.4 (8.0; 23.1) | 0.063 ² |

¹Paired t-test. ²Wilcoxon test.

^aFive children received calcium supplementation on the day of the oral food challenge and after 30 days (median = 400.0 mg, 25th and 75th percentiles: 212.0; 500.0). ^b40 children received iron supplementation on the day of the oral food challenge (median = 10.0 mg, 25th and 75th percentiles: 9.0; 23.5), and 36 children received it after 30 days (median = 10.0, 25th and 75th percentiles: 8.5; 22.5).

^c46 children received vitamin A supplementation on the day of the oral food challenge (median = 750.0 µg, 25th and 75th percentiles: 750.0; 750.0), and 44 children received it after 30 days (median = 750.0 µg, 25th and 75th percentiles: 750.0; 750.0). ^dNine children received vitamin C supplementation on the day of the oral food challenge and after 30 days (median 40.0 mg, 25th and 75th percentiles: 30.0; 110.0). ^e47 children received vitamin D supplementation on the day of the oral food challenge (median = 12.5 mg, 25th and 75th percentiles: 12.5; 12.5), and 45 children received it after 30 days (median = 12.5 mg, 25th and 75th percentiles: 12.5; 12.5).

the test. The mineral and vitamin supplements were maintained at the same dose after the OFC.

Adequate intake of macro- and micronutrients relative to DRIs

Table 3 shows the frequency (number and percentages) of children who had adequate intake of macro- and micronutrients relative to DRIs before and after the OFC. There was a statistically significant increase in the proportion of children with adequate calcium and phosphorus intake.

Nutritional status

Table 4 provides the Z-scores for weight-for-age, weight-for-height, height-for-age, and BMI-for-age on the day of the oral food challenge and after 30 days. There was a statistically significant increase in the weight-for-age and height-for-age Z-scores. The weight-for-height and BMI-for-age Z-scores showed no statistically significant variation.

Discussion

The increase in children's food intake after discontinuing the cow's milk protein-free diet showed that the return to the usual diet, that is, without restrictions, was associated with an improvement in the children's food intake, including the proportion of adequate intake of calcium in relation to DRIs. It is likely that the increase in food intake is related to the possibility of using greater variety of foods

in the diet, especially cow's milk and other foods containing cow's milk proteins. There was also an increase in weight-for-age and height-for-age Z-scores.

These results reinforce the concept that an elimination diet should be used only for the period that is strictly necessary. Therefore, it is essential to perform OFC not only to confirm the diagnosis of CMA but also to characterize the development of oral tolerance. In our sample, of the 130 children who underwent the oral challenge test, 101 (77.6%) had a negative OFC, that is, they changed to a diet without any restrictions for cow's milk and dairy products. A similar rate of negativity for the oral challenge test was observed in a previous study.¹¹ Conversely, other studies conducted in Recife (Brazil)¹² and the Netherlands³² found a lower positivity rate of approximately 50.0%. This difference was likely observed because in our sample, the oral challenge test was performed predominantly for the study of oral tolerance, while in the aforementioned studies, the challenge food was used predominantly to establish the diagnosis of CMA.^{11,32} In other words, the expectation of positivity, in general, is higher when the test is for diagnostic purposes rather than for oral tolerance assessments.

According to international consensus, it is important to systematically monitor the development of oral tolerance by children with food allergies. Therefore, OFC should be performed every 6-12 months whenever possible to identify oral food tolerance.^{1,3,8}

Regarding the use of special formulas, our study showed that extensively hydrolyzed formula and amino acid formula were the most commonly used. Notably, a recent study in Brazil³³ showed that special formulas (based on extensively hydrolyzed protein and amino acids) represent an important source of nutrients for infants on

Table 3 Frequency (number and percentages) of children with adequate and inadequate energy, protein, carbohydrate, lipids, phosphorus, calcium, iron, and vitamins A, C, and D intake through food, based on dietary reference intakes, on the day of the oral food challenge and after 30 days

| | | After 30 days | | | | | | P ¹ |
|--------------------------------|------------|---------------|--------|----------|---------|-------|---------|----------------|
| | | Inadequate | | Adequate | | Total | | |
| | | n | % | n | % | n | % | |
| Energy | | | | | | | | |
| On the day | Inadequate | 26 | (32.5) | 12 | (15.0) | 38 | (47.5) | 0.146 |
| | Adequate | 5 | (6.3) | 37 | (46.2) | 42 | (52.5) | |
| | Total | 31 | (38.8) | 49 | (61.2) | 80 | (100.0) | |
| Protein | | | | | | | | |
| On the day | Inadequate | 0 | (0.0) | 4 | (5.0) | 4 | (5.0) | 0.134 |
| | Adequate | 0 | (0.0) | 76 | (95.0) | 76 | (95.0) | |
| | Total | 0 | (0.0) | 80 | (100.0) | 80 | (100.0) | |
| Carbohydrate | | | | | | | | |
| On the day | Inadequate | 13 | (16.3) | 15 | (18.7) | 28 | (35.0) | 0.136 |
| | Adequate | 7 | (8.7) | 45 | (56.3) | 52 | (65.0) | |
| | Total | 20 | (25.0) | 60 | (75.0) | 80 | (100.0) | |
| Lipids | | | | | | | | |
| On the day | Inadequate | 22 | (27.5) | 14 | (17.5) | 36 | (45.0) | 0.540 |
| | Adequate | 10 | (12.5) | 34 | (42.5) | 44 | (55.0) | |
| | Total | 32 | (40.0) | 48 | (60.0) | 80 | (100.0) | |
| Phosphorus | | | | | | | | |
| On the day | Inadequate | 7 | (8.7) | 14 | (17.5) | 21 | (26.3) | 0.022 |
| | Adequate | 4 | (5.0) | 55 | (68.8) | 59 | (73.7) | |
| | Total | 11 | (13.7) | 69 | (86.3) | 80 | (100.0) | |
| Calcium | | | | | | | | |
| On the day | Inadequate | 23 | (28.7) | 22 | (27.5) | 45 | (56.3) | <0.001 |
| | Adequate | 4 | (5.0) | 31 | (38.8) | 35 | (43.7) | |
| | Total | 27 | (33.7) | 53 | (66.3) | 80 | (100.0) | |
| Calcium + supplements | | | | | | | | |
| On the day | Inadequate | 22 | (27.5) | 18 | (22.5) | 40 | (50.0) | <0.012 |
| | Adequate | 5 | (6.2) | 35 | (43.8) | 40 | (50.0) | |
| | Total | 27 | (33.7) | 53 | (66.3) | 80 | (100.0) | |
| Iron | | | | | | | | |
| On the day | Inadequate | 16 | (20.0) | 9 | (11.3) | 25 | (31.3) | 0.170 |
| | Adequate | 17 | (21.2) | 38 | (47.5) | 55 | (68.7) | |
| | Total | 33 | (41.2) | 47 | (58.8) | 80 | (100.0) | |
| Iron + supplements | | | | | | | | |
| On the day | Inadequate | 4 | (5.0) | 4 | (5.0) | 8 | (10.0) | 0.121 |
| | Adequate | 11 | (13.8) | 61 | (76.2) | 72 | (90.0) | |
| | Total | 15 | (18.8) | 65 | (81.2) | 80 | (100.0) | |
| Vitamin A | | | | | | | | |
| On the day | Inadequate | 7 | (8.7) | 11 | (13.8) | 18 | (22.5) | 0.831 |
| | Adequate | 11 | (13.8) | 51 | (63.7) | 62 | (77.5) | |
| | Total | 18 | (22.5) | 55 | (77.5) | 80 | (100.0) | |
| Vitamin A + supplements | | | | | | | | |
| On the day | Inadequate | 4 | (5.0) | 6 | (7.5) | 10 | (12.5) | 1.000 |
| | Adequate | 5 | (6.3) | 65 | (81.2) | 70 | (87.5) | |
| | Total | 9 | (11.3) | 71 | (88.7) | 80 | (100.0) | |
| Vitamin C | | | | | | | | |
| On the day | Inadequate | 5 | (6.2) | 3 | (3.8) | 8 | (10.0) | 0.505 |
| | Adequate | 6 | (7.5) | 66 | (82.5) | 72 | (90.0) | |
| | Total | 11 | (13.7) | 69 | (86.3) | 80 | (100.0) | |
| Vitamin C + supplements | | | | | | | | |
| On the day | Inadequate | 3 | (3.8) | 3 | (3.8) | 6 | (7.5) | 0.343 |
| | Adequate | 7 | (8.7) | 67 | (83.7) | 74 | (92.5) | |
| | Total | 10 | (12.5) | 70 | (87.5) | 80 | (100.0) | |

(Continued)

Table 3 (Continued)

| | | After 30 days | | | | | | P ¹ |
|-------------------------|------------|---------------|--------|----------|--------|-------|---------|----------------|
| | | Inadequate | | Adequate | | Total | | |
| | | n | % | n | % | n | % | |
| Vitamin D | | | | | | | | |
| On the day | Inadequate | 44 | (55.0) | 12 | (15.0) | 56 | (70.0) | 0.080 |
| | Adequate | 5 | (6.3) | 19 | (23.7) | 24 | (30.0) | |
| | Total | 49 | (61.3) | 31 | (38.7) | 80 | (100.0) | |
| Vitamin D + supplements | | | | | | | | |
| On the day | Inadequate | 28 | (35.0) | 8 | (10.0) | 36 | (45.0) | 1.000 |
| | Adequate | 7 | (8.7) | 37 | (46.3) | 44 | (55.0) | |
| | Total | 35 | (43.7) | 45 | (56.3) | 80 | (100.0) | |

¹McNemar test.**Table 4** Z-scores for weight and height and frequency of the anthropometric deficit on the day of oral food challenge and after 30 days.

| Variable | On the day (n = 76) | After 30 days (n = 76) | P |
|--|------------------------|------------------------------|---------------------|
| Z-scores | | | |
| Weight-for-age | -0.6 (-1.7; 0.0) | -0.4 (-1.4; 0.2) | <0.001 ¹ |
| Weight-for-height | -0.1 ± 1.1 | -0.1 ± 1.0 | 0.793 ² |
| Height-for-age | -1.1 ± 1.6 | -0.8 ± 1.4 | <0.001 ² |
| Body mass index-for-age | -0.0 ± 1.1 | -0.0 ± 0.9 | 0.961 ² |
| Anthropometric deficit (Z-scores < -2.0) | | | |
| Weight-for-age | 12 (15.8%) | 9 (11.8%) | 0.248 ³ |
| Height-for-age | 18 (23.7%) | 14 (18.4%) | 0.182 ³ |
| Weight-for-height | 3 (3.9) | 3 (4.0%) | 0.480 ³ |
| Body mass index-for-age | 2 (2.6%) | 1 (1.3%) | 1.000 ³ |

¹Wilcoxon test; ²Paired *t* test; ³McNemar test.

an elimination diet even after the introduction of complementary feeding.

Our study showed relevant information regarding calcium intake. Although most patients were being monitored in a multidisciplinary unit, a portion of patients (56.3%) had inadequate calcium intake, even when receiving supplementation. A study¹⁷ conducted in Italy showed that nutrition counseling was accompanied by a higher intake of energy, carbohydrates, proteins, and calcium. Nutritional monitoring is important during the cow's milk elimination diet. Counseling by a pediatrician and dietician experienced in pediatric nutrition is highly recommended to ensure adequate nutrient intake and monitor the nutritional status of patients.^{1,8,17,34} In this study, the impact of nutritional guidance was not evaluated. Previously, it was observed in Brazil that there may be limitations in the understanding of patients regarding nutritional guidelines and their ability to implement dietary recommendations.³⁵ Based on these findings, the care protocols and guideline adherence protocols should be reassessed.

Our study showed that discontinuation of the cow's milk protein elimination diet was accompanied by higher calcium intake and a higher proportion of children with an adequate diet relative to DRIs for most macro- and micronutrients, with the maintenance of the same doses of mineral and vitamin supplements. Our findings before discontinuing the cow's milk protein-free diet are in agreement with previous studies^{13,21} that found nutrient deficits, especially for energy, proteins, lipids, and calcium, in the diet of children with CMA relative to the DRIs and relative to the food intake of healthy children. Thus, the dietary restriction required to control CMA can negatively impact food intake and impair child growth and development.^{11,13,18,22}

In addition to the changes in food intake, there was an impact on anthropometric parameters, that is, 30 days after discontinuation of a diet free of cow's milk and dairy products, there were fewer children with Z-scores < -2.0 standard deviations for weight-for-age, height-for-age, and BMI-for-age; however, the variation was not statistically significant. On the other hand, there was a statistically significant increase in the individual weight-for-age and height-for-age Z-scores. Importantly, on the day of the OFC, the predominant anthropometric deficit was height-for-age, found in 23.7% of children, similar to that observed in a previous study conducted in Brazil.⁴ Other studies in Brazil^{13,16} and in other countries^{17,18,19,36,37} reported lower height-for-age, weight-for-age and Weight-for-height Z-scores for children on a cow's milk protein elimination diet than for healthy children.

As already mentioned, only one study in the literature has evaluated the impact of diet discontinuation on growth.²² Only the height measurements of 58 children who had a negative OFC and another height measurement approximately 1 year after discontinuing the cow's milk protein elimination diet were retrospectively evaluated. The authors observed, relative to the Japanese growth curve, an increase in height-for-age Z-scores from -0.28 ± 0.96 , at the time of diet discontinuation, to -0.12 ± 1.07 , 1 year later ($P = 0.017$). In our study, there was an increase not only in height-for-age but also in weight-for-age observed in a period of only 30 days. Despite these variations in the individual Z-scores, there was no reduction in the percentage of children with anthropometric deficits, that is, with Z-scores less than -2.0 standard deviations. It is interesting

to note that in the Japanese study, although the authors do not provide information on food intake, the possible increase in calcium intake was considered the most likely explanation for the improvement in height-for-age Z-score. In our study, there was an increase in both calcium intake and a reduction in the proportion of children with intake below the DRIs. There are many factors associated with the growth and development of children with CMA, such as food intake, inflammation during the allergic process, and nutrient bioavailability. However, this issue should be evaluated in future studies.

The present study has some limitations, such as the lack of a control group. However, it would not be ethical to keep children with a negative OFC on an elimination diet. In addition, the 30-day follow-up period can be considered short; however, it was sufficient to show statistically significant variations not only in feeding but also in anthropometric parameters.

Conclusion

Our study showed that after a negative OFC, the inclusion of cow's milk and dairy products in the diet was associated with an increase in the intake of energy, proteins, carbohydrates, calcium, phosphorus, and vitamin D and increased weight-for-age and height-for-age Z-scores. These findings are an incentive to perform OFC in order to discontinue elimination diets at the most appropriate time.

Acknowledgments

This study was supported by National Council for Scientific and Technological Development (Conselho Nacional de Desenvolvimento Científico e Tecnológico - CNPq).

Conflict of Interest

The authors declare no potential conflicts of interest with respect to research, authorship, and/or publication of this article.

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