

## Dietary intake of protein sources in infants between 7-24 months old: start time and compliance with recommendations

### Ingesta alimentaria de fuentes proteicas en lactantes entre 7-24 meses: tiempo de inicio y cumplimiento de recomendaciones

Natalia Sepúlveda Alarcón<sup>a</sup>, Catalina Le Roy Olivos<sup>b</sup>

<sup>a</sup>Residente, Programa de Especialización en Pediatría, Facultad de Medicina Pontificia Universidad Católica de Chile. Santiago, Chile

<sup>b</sup>Departamento de Gastroenterología y Nutrición Pediátrica, Facultad de Medicina Pontificia Universidad Católica de Chile. Santiago, Chile

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#### What do we know about the subject matter of this study?

According to the 2010 national food consumption survey, there is low compliance with the food consumption recommendations of the national guidelines, especially in fish and legumes; however, such survey only included children over 24 months.

#### What does this study contribute to what is already known?

This study provides information on compliance with dietary recommendations in infants between 7-24 months, finding that the incorporation of legumes, eggs, and fish is late, and the consumption of other meats is excessive, similar to the rest of the Chilean population.

#### Abstract

Adequate nutrition includes breastfeeding, infant formula, and the incorporation of complementary feeding (CF). **Objective:** To describe compliance with the recommendations for protein intake from CF in healthy infants between 7 and 24 months old. **Patients and Method:** Healthy infants from an outpatient center were studied. To collect the variables under study, we designed and applied a questionnaire. We obtained demographic data, breastfeeding, use of formula, and incorporation and compliance with protein intake data. The analysis of variables was carried out with STATA software version 13, and the Chi-square test and one-way ANOVA were applied. **Results:** 85 infants were studied, 54% were female, 68% of infants were exclusively breastfed until 6 months old, and 84.7% started complementary feeding at that age. 37.6% complied with the incorporation of fish, 49.4% with legumes, and 45.8% with egg. 52% of infants aged between 10 and 12 months and 83% of infants between 13 and 24 months consumed more beef, chicken, or turkey. This data was associated with being the first child and older age, as well as egg consumption where infants older than 12 months ate more than recommended. We observed lower than recommended fish consumption in 78% of infants aged between 10 and 12 months and 83.2% of infants between 13 and 24 months, of which 35% and 2.7%

#### Keywords:

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Correspondence:  
Catalina Le Roy  
catalinaleroy@yahoo.es

had never eaten fish, respectively. 45 infants were fed only with formula and complementary feeding, 28 (62%) of them received  $\geq 1$  g/kg/day of protein from infant formula. **Conclusions:** Most infants were exclusively breastfed until 6 months old and incorporated complementary feeding at that age. Most infants incorporated legumes, eggs, and fish later than recommended, and there was excessive consumption of beef, chicken, or turkey and little consumption of fish, especially at older ages.

## Introduction

During the first two years of life, there is a transition from exclusive dairy feeding to mixed feeding based on dairy and solid foods, which are incorporated into the family table<sup>1</sup>. The main protein sources during this period are dairy (breast milk (BM) and/or milk formulas (MF)), meats (beef, chicken, turkey, pork, fish), legumes, and eggs<sup>1,2</sup>.

The intake recommendations are to avoid deficits or excesses of proteins and other important nutrients in this stage of rapid growth, naturally contained in these protein sources or dairy products such as iron, zinc, vitamin B12, and essential fatty acids<sup>3</sup>.

In 2010, the National Food Consumption Survey (*Encuesta Nacional Consumo Alimentario- ENCA*) was conducted, which included the population above years-old of age. Nationally, only 13.7% complied with the consumption recommendations of the national guidelines, where fish, dairy products, and legumes were those with the lowest compliance; highlighting the group between 2 and 5 years of age, that presented a low intake of fish (21.5%) and legumes (31%)<sup>4</sup>.

The association between protein intake and excess weight is currently being studied, and it has been found that infants under one year of age fed with MF with a higher protein content than BM have a higher risk of presenting obesity at school age, however, this is not yet clear for the second year of life<sup>5,6</sup>.

The main objective of this study is to describe the compliance with the recommendations of the current national guidelines for normal feeding<sup>1</sup>, regarding the incorporation of different protein sources and their frequency of consumption in healthy infants aged between 7 and 24 months.

## Patients and Method

Cross-sectional study with prospective selection. Healthy infants aged 7 to 24 months evaluated at the San Joaquín outpatient care center of the UC Christus Health Network, between February 2019 and March 2020, were included. Children were excluded if they

had any pathology affecting or restricting their feeding such as food allergy, use of nasogastric tube or gastrostomy, renal or hepatic insufficiency, metabolic diseases, vegetarian options, premature infants, and children of immigrants who can maintain feeding schemes of their nationality.

We developed questionnaires to obtain the following information: date of birth, sex, weeks of gestation and birth weight, nursery attendance, birth order, responsible for feeding, extended family living in the same household (relatives other than parents and siblings), age and maternal schooling. The most recent weight and height were obtained from the electronic health record and the nutritional diagnosis was made according to WHO growth standards and ministerial definitions<sup>7</sup>.

We asked about the duration of exclusive breastfeeding (EBF), total duration of EBF, age of incorporation of MF, MF used, amount of formula consumed per day, and its preparation. The formula used was classified as first infant formula, follow on formula, National Complementary Feeding Program (Programa Nacional de Alimentación Complementaria - PNAC) formula for children under 18 months (Purita fortified), commercial formula for children over 1 year of age, PNAC formula for children over 18 months (Purita Cereal), and other formulas that contained some specific change in their components. In the case of infants using only MF at the time of the study, the g/kg/day of protein consumed was calculated.

Table 1 shows the complementary feeding (CF) recommendations according to the 2016 national feeding guideline. Based on this guideline, three groups were defined: group I (GI) between 7-9 months, group II (GII) 10-12, and group III (GIII) 13-24 months, according to the recommended age to start intake of each protein source (beef, chicken, turkey, pork, fish, legumes, egg) and the frequency to consume each of them in the week<sup>1</sup>. In this way, it was determined whether the foods were incorporated appropriately, early or late, and whether they complied with the recommended weekly frequency of consumption of each one of them.

Although this study can be considered exploratory, we calculated the sample size based on the number of

children included in the ENCA, which had a sample of 257 surveyed children aged 2 to 5 years, subdivided into 3 groups according to age, resulting in 85 subjects/year; therefore, by studying around only one age group, we calculated a total of 85 subjects, who were selected prospectively, completing a similar number among the three defined groups.

Data were analyzed using STATA software version 13, and categorical variables were described as relative and absolute frequency. The Chi-square test was used for associations of categorization of start and compliance with feeding recommendations with maternal age greater or less than 30 years, protein intake from MF greater or less than 1 g/kg/day, birth weight for gestational age, maternal schooling, birth order, nursery attendance, and the person responsible for food preparation. One way ANOVA was performed to compare the g/kg/day of protein from MF among the 3 age groups. A value  $p < 0.05$  was considered statistically significant.

This research was approved by the ethics committee of the Faculty of Medicine of the Pontifical Catholic University of Chile, number 180322008. Written informed consent was obtained from the parents before study entry.

## Results

85 subjects were studied. Table 2 shows their data and demographic variables. It should be noted that 54% were female, none had short height, 54% were the first child, and in 70% of them, the mother was responsible for feeding. 68% received EBF up to 6 months of age, 54.9% of those older than 12 months were still receiving BM, 44.6% started CF at 6 months or earlier, and 21% after one year of age. Of the infants fed only with MF plus CF, 62% received  $\geq 1$  g/kg/day of protein from MF. Table 3 shows the milk intake according to the study groups, presenting no significant difference in the average intake of grams of protein/kg/day from MF ( $p = 0.2$ ).

Out of the study group, 84.7% started CF at 6 months, 90.5% started intake of beef, chicken, turkey, or pork at 7 months, 37.6% fish at 8 months, 49.4% legumes at 9 months, and 45.8% eggs at 11 months. Figure 1 shows the early, late, adequate, or no incorporation of each protein source. Those that were not yet of the recommended age for incorporation are represented as “not applicable”.

22/26 of the GI, 18/23 of the GII, and 21/36 of the GIII complied with the recommendation to start eating beef, chicken, turkey, or pork at 6 months. Regarding fish, 4/26 of the GI, 3/23 of the GII, and 6/36 of the GIII complied with the recommendation to start eating

fish. It is worth mentioning that 21/26 of the GI and 8/23 of the GII had never eaten fish.

On the other hand, 6/26 of the GI, 10/23 of the GII, 13/36 of the GIII complied with legume incorporation. It should be noted that 5/26 infants from the GI were not old enough to incorporate legumes, therefore they were not considered in the compliance item, while 15/21 were old enough to incorporate legumes and had not yet done so. In the GIII, 7/36 incorporated legumes early and 16/36 did so late.

9/23 of GII and 13/36 of GIII met the incorporation of egg. 6/23 of GII and 9/36 of GIII incorporated egg early, while 3/26 of GII and 16/36 of GIII did so late. 5/23 of GII had never eaten eggs. Figure 2 shows compliance with the frequency of consumption of each food for groups II and III.

Searching for an association between variables of weight adequacy for gestational age, maternal age, maternal schooling, nursery attendance, child's nutritional diagnosis, age group, birth order, duration of EBF, total BF, start of MF with the start of mashed CF, and start and compliance with the recommendations for each of the protein sources studied (beef, chicken, turkey, pork, legumes, fish, and eggs) we found that those who were the first child consumed more meat than recommended ( $p = 0.01$ ). This was also observed in the older age groups (10 to 24 months) ( $p < 0.001$ ), where those older than 12 months ate more eggs than recommended ( $p < 0.001$ ), GII had a lower intake of legumes than recommended ( $p < 0.001$ ) and in all groups, fish intake was the lowest ( $p < 0.001$ ). There was no association between nutritional diagnosis with the dairy source used or the g/kg/day of MF protein, in those where the calculation was applicable.

It was found that those who did not attend nursery complied more frequently with the recommendation for legume intake ( $p = 0.006$ ).

**Table 1. Recommendations for the incorporation and weekly consumption of protein sources in complementary feeding infants<sup>1</sup>**

Food	Start (month)	Recommendations (times/week)		
		GI (7-9 month)	GII (10-12 month)	GIII (13-24 month)
Meats*	6	3-7	3	2
Fish	6-7	0-2	2	2
Legumes	7-8	0-2	2	1-2
Egg	9-10	0-2	0-2	1-2

\*Meats: beef or chicken or turkey or pork.

**Table 2. Variables and demographic data of infants studied**

Variable	Frecuency n (%)
Age	
Group I	26 (31)
Group II	23 (27)
Group III	36 (42)
Sex	
Female	46 (54)
Male	39 (46)
Adequacy BW	
SGA	6 (7)
AGA	68 (80)
LGA	11 (13)
Nutritional Diagnosis	
Risk of malnutrition	2 (3)
Eutrophy	47 (61)
Overweight	17 (22)
Obesity	11 (14)
Maternal Age	
20-30 years	35 (41)
31-43 years	50 (59)
Son order	
First	46 (55)
Second	31 (36)
Third	8 (9)
Nursery attendance	
Not	66 (78)
Yes	19 (22)
Extended family	
No	62 (73)
Yes	23 (27)
Responsible for feeding	
Mother	60 (71)
Grandparents	11 (13)
Home advisor	1 (1)
Another close relative	1 (1)
Nursery	12 (14)
Maternal education	
Completed secondary	18 (21)
Complete higher education	67 (78)

BW: birth weight; GA: Gestational age; SGA: small for gestational age; AGA: appropriate for gestational age; LGA: large for gestational age.

## Discussion

Feeding in the first 1,000 days of life is one of the main determinants of growth and development in infancy<sup>8</sup>. During this period, infants receive protein from different sources contained in BM, MF, and CF<sup>6</sup>.

BM is recommended up to 6 months due to its benefits for both the child and the mother<sup>9</sup>. Between 2005 y 2014 in Chile, the prevalence of EBF at 6 months was 51%<sup>10</sup>. In our study, a large percentage of patients had EBF up to 6 months, showing no significant associations between maternal schooling and compliance with this recommendation, unlike a study conducted in 2012 in our country, where it was found that, among other factors, lower maternal schooling was more related to EBF up to 6 months<sup>11</sup>.

Since BM provides energy and immunological factors during the first two years of age, it is recommended to extend it to two years of life or more, as long as adequate CF and good nutritional status are maintained<sup>2</sup>. Out of the infants studied over one year of age, more than half received BM.

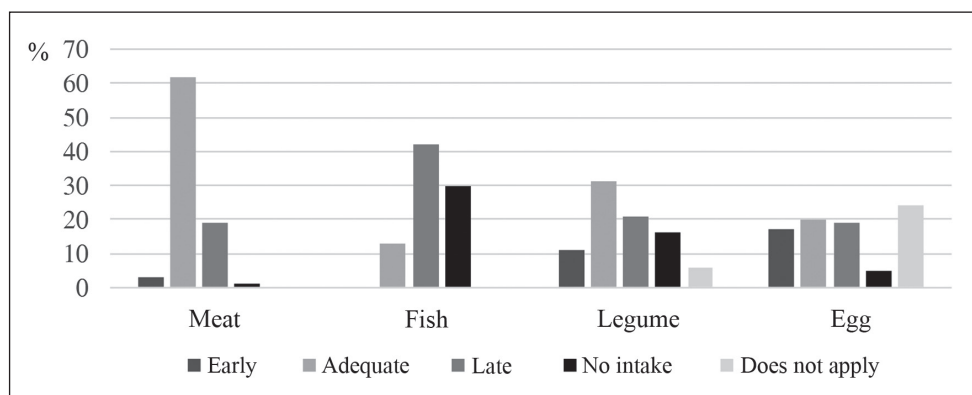
It is known that the nutrients provided by BM are different from those provided by MF, especially in the amount of protein<sup>3</sup>. There has been an association between excess weight at school and feeding with MF with higher protein content in the first year of life<sup>5,12</sup>, and on the other hand, a systematic review, based on 4 randomized controlled studies, concludes that the use of MF with low protein concentration (similar to BM) results in an adequate increase in weight and height<sup>13</sup>. Although in our study there was a high percentage of infants fed with BM, more than half of them exclusively used first infant or follow on formula or continuation formula for their dairy feeding, however, there was no association with excess weight.

When quantifying protein intake by MF, we found that a large percentage received  $\geq 1$  g/kg/day of protein from this source alone, exceeding the WHO recom-

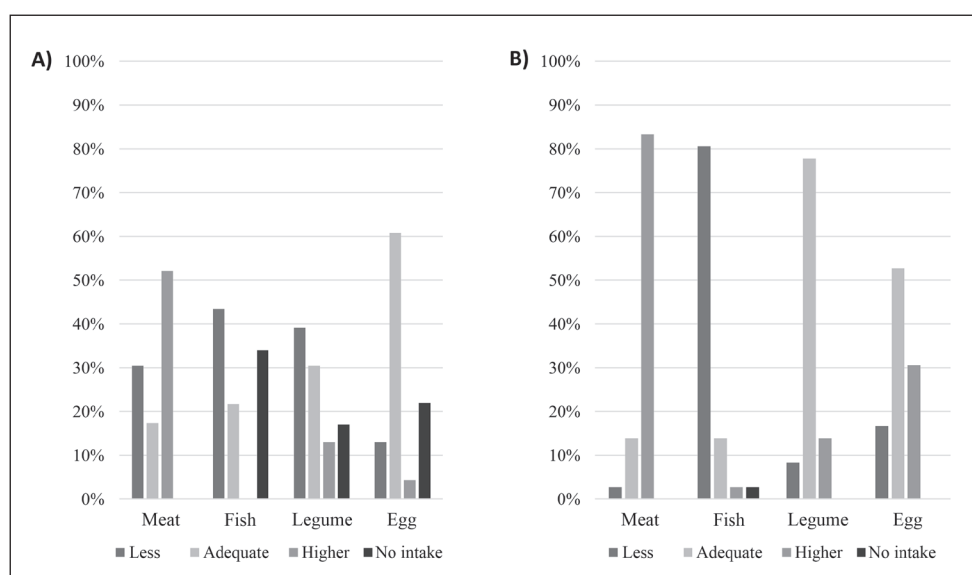
**Table 3. Frecuency and percentage of dairy feeding according to study group**

Dairy feed	GI n = 26	GII n = 23	GIII n = 36
Breast milk (%)	13 (50)	7 (30.4)	5 (13.8)
Mixed (BM + MF), n (%)	2 (7.6)	2 (8.6)	11 (30.5)
Use of formula			
First infant formula, n (%)	2 (7.6)	1 (4.34)	
Follow on formula, n (%)	3 (11.5)	3 (13.0)	1 (2.7)
PNAC, n (%)	2 (7.6)	4 (17.3)	7 (19.4)
Commercial formula for > 1 year, n (%)	1 (3.8)	4 (17.3)	12 (33.3)
Other, n (%)	3 (11.5)	2 (8.6)	
g protein/kg/day* (mean, DS)	1.3 $\pm$ 0.8	1.15 $\pm$ 0.6	0.9 $\pm$ 0.4

\*Calculated only for those infants with milk formula and without breast milk. BM: breast milk; MF: milk formula; PNAC: National complementary feeding program.



**Figure 1.** Compliance with the recommendation for the incorporation of protein sources in complementary infant feeding (n = 85).



**Figure 2.** Compliance with recommendation frequency of intake of protein sources in complementary feeding of infants.

**A:** Group II (10-12 month, n = 23);  
**B:** Group III (13-24 month, n = 36).

mendation, which establishes 1 g/kg/day as a safe level of protein intake that should be provided by different sources. We did not find significant associations between this protein intake and nutritional diagnosis, so we recommend a study with a further follow-up of the subjects; however, based on the evidence, studies are currently underway to evaluate whether the amount of protein in milk formula can be safely reduced<sup>3</sup>. Although several studies positively associate the amount of MF protein with greater weight gain and higher BMI in the long term, the effect of higher protein intake from different sources (dairy plus CF) is not clear, with mixed results<sup>6,14</sup>.

From 6 months of age, CF should be initiated since BM does not cover all the needs of the child, mainly the intake of micronutrients such as iron, zinc, and essential fatty acids<sup>3</sup>. A Mexican study found that the average age of initiation of CF was 6.1 months<sup>15</sup>, which is consistent with our results, and a statistically significant association was found with a eutrophy as nutritional diagnosis, in contrast, a study conducted in

8 European countries showed that infants who initiated CF  $\geq 7$  months had a higher prevalence of excess weight in infancy<sup>16</sup>.

During the last few years, there has been a great change in the incorporation of the foods of this study because, despite the late introduction of potentially allergenic foods, the prevalence of food allergy continues to increase<sup>17</sup>. Studies showed that delaying the introduction of such foods could increase the risk of IgE sensitization and food allergy<sup>18</sup>. Evidence now suggests that the early introduction of these foods is an effective preventive strategy<sup>19</sup>. A 2016 meta-analysis including 6 studies, concludes that early introduction of egg (6 months) is associated with a lower risk of egg allergy<sup>20</sup>.

It will take some time for the population to incorporate this type of food from 7 months of age; moreover, we found that more than half of the population incorporated eggs, fish, and legumes late. We found no association between late incorporation of these foods with maternal age or with the person responsible for infant feeding.

In both GII and GIII, there is low compliance with the frequency of fish consumption, higher meat intake, especially in the GIII group, where the results are similar to those found in the 2 to 5 year-old group and in the general Chilean population of the ENCA, probably due to the incorporation of the child to the family table<sup>4</sup>.

Regarding dietary variability, studies in South Asia identified lack of maternal education and lower socioeconomic status as determinants of inadequate CF practices<sup>21</sup>, which is consistent with the findings of the ENCA in our country, which showed that the high socioeconomic status has a more varied diet with higher consumption of fish and meat, while the low one has a higher consumption of legumes and low consumption of fish. Although we did not study the socioeconomic level, the low compliance with the fish recommendation stands out, both in the introduction of fish at 8 months and in the frequency of weekly consumption.

Among the limitations of our study, we consider that these results cannot be generalized to the total Chilean population; however, they constitute a first approximation to this age group. Another limitation is that, as this is an intake trend study, it is not possible to measure the total protein intake of the child.

Our results are a first contribution to describe the dietary intake of infants, which would allow us to identify the points to emphasize in the feeding recommendations in the daily practice of our well-baby checkups.

## Conclusions

In the studied population, most of the patients comply with the EBF recommendation at 6 months and the beginning of CF. The incorporation of legumes, eggs, and above all fish, in general, is late and their consumption is lower than recommended. In contrast, the consumption of beef, chicken, turkey, or

pork is higher than recommended, which is similar to the general Chilean population, as well as to the 2 to 5 year-old group that has been studied in Chile. There were some associations between the variables studied and the initiation of and compliance with the intake recommendations, so it is important to continue studying them with more representative groups in our country and relating them to some long-term effects such as allergic diseases and excess weight.

## Ethical Responsibilities

**Human Beings and animals protection:** Disclosure the authors state that the procedures were followed according to the Declaration of Helsinki and the World Medical Association regarding human experimentation developed for the medical community.

**Data confidentiality:** The authors state that they have followed the protocols of their Center and Local regulations on the publication of patient data.

**Rights to privacy and informed consent:** The authors have obtained the informed consent of the patients and/or subjects referred to in the article. This document is in the possession of the correspondence author.

## Conflicts of Interest

Authors declare no conflict of interest regarding the present study.

## Financial Disclosure

Authors state that no economic support has been associated with the present study.

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