





Original/Pediatría

Development and validation of two food frequency questionnaires to assess gluten intake in children up to 36 months of age

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Abstract

Background and objective: precise information on gluten consumption is crucial for specifically studying the impact of gluten introduction and gluten intake in celiac disease development. Our aim was to develop and validate tools (food frequency questionnaires, FFOs) for the assessment of gluten consumption in Spanish children aged 7-36 months.

Methods: a total of 342 children, who attended primary healthcare centers for routine health surveys or La Fe Hospital for minor health problems as well as healthy children (recruited in nurseries and primary schools) participated in this survey. We have developed two different FFQs (one for 7-12 months and other for 13-36 months). For validation, results from two FFQs were compared with results of 2-day food records and also with the gold standard 7-day records. The mean gluten intake obtained by the 2DR vs. FFQ and the 7DR vs. FFQ, were compared using the Bland Altman plot method and also Lin's concordance correlation coefficient.

Results: we found a good agreement between our FFQs and the 2DR and 7DR according to the results of both the Bland-Altman plots and Lin's concordance correlation coefficient.

Conclusions: our two new FFOs are therefore the only validated questionnaires available to determine gluten consumption in Spanish children. They are user-friendly and offer excellent instruments to assess gluten intake in children up to 36 months of age.

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Key words: Celiac disease. Gluten. Food frequency questionnaire. Spanish. Children.

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DESARROLLO Y VALIDACIÓN DE DOS CUESTIONARIOS DE FRECUENCIA DE CONSUMO PARA EVALUAR LA INGESTA DE GLUTEN EN NIÑOS HASTA LOS 3 AÑOS

Resumen

Antecedentes y objetivos: una información precisa sobre el consumo de gluten es muy importante para estudiar el verdadero impacto de la introducción y la ingesta de gluten en el desarrollo de la enfermedad celiaca. El objetivo del estudio fue desarrollar y validar herramientas (cuestionarios de frecuencia de consumo, CFC) para evaluar el consumo de gluten en niños con edades comprendidas entre los 7 y 36 meses.

Métodos: se incluyeron un total de 342 niños reclutados en el Hospital Universitario y Politécnico de La Fe, así como en guarderías y escuelas de primaria con problemas menores de salud. Se desarrollaron dos CFC diferentes (uno para niños de 7 a 12 meses y otro para niños de 13 a 36 meses). Para su validación, los resultados obtenidos con los CFC se compararon con un registro alimentario de dos días (2RA) y también con un registro alimentario de soete días (7RA), considerado el "estándar de oro". La ingesta media de gluten obtenida de la comparación del 2RA vs. CFC y de 7RA vs. CFC fueron comparadas usando el método de Bland Altman plot y también el coeficiente de correlación de concordancia de Lin's.

Resultados: de acuerdo a los resultados de los dos métodos estadísticos usados para la validación, se encontró una buena correlación entre los CFC y los registros alimentarios de dos y siete días, lo que indica que los CFC son fiables para la evaluación de la ingesta de gluten.

Conclusiones: estos dos nuevos CFC son los únicos validados y disponibles en España para la evaluación de la ingesta de gluten. Además, son herramientas útiles y fáciles de usar para el cálculo del consumo de gluten en niños de hasta tres años de edad.

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Palabras clave: Enfermedad celiaca. Gluten. Cuestionario de frecuencia de consumo. Español. Niños.

Introduction

Celiac disease (CD) is an inflammatory intestinal disorder caused by immune responses, an abnormal immune response induced by dietary gluten proteins1. Currently, it is the most common food intolerance in Europe with a minimum prevalence estimated of 1/100 individuals². A strong association with certain HLA antigens has been verified. Most CD patients are HLA-DQ2-positive, whereas the remainders are usually HLA-DQ8-positive. However, less than 1/2 of genetically susceptible individuals develop CD, even though virtually all individuals in wheat-consuming populations are exposed to gluten. This suggests that additional factors play a role in disease risk²⁻⁴. Classics studies suggest that the timing of gluten introduction as well as the amount of gluten in relation to breast-feeding could be related to a higher risk of CD⁵⁻⁹. To confirm these hypothesis, specific studies have been designed, for instance Prevent CD (http:// www.preventceliacdisease.com), which aims to establish the exact amount of gluten that children consume and to link it with other possible interplaying co-factors¹⁰.

It is not easy to precisely determine what an individual eats. Nonetheless, there are various methods available for dietary assessment done by means of in-taking estimations. One of them is the food record (FR). which consists off keeping accurate records of all the food items consumed within specific days, and it can be considered an estimate of an individual's daily intake. Another method is the food-frequency questionnaire (FFQ), which estimates the frequency of food consumed per week. Currently, the FFQ is the most commonly used method in industrialized countries to assess food intake in large epidemiologic studies, and also to investigate the relationship between habitual diet and disease¹¹⁻¹³. FFOs are considered appropriate tools for measuring energy and macronutrient consumption in several countries. These are reasonable tools to determine micronutrient intake. However, it has been argued that FFQs may need specific validation for each region in order to be culturally sensitive and to correspond to the prevailing food culture^{11,14}.

There are different validated FFQs in order to assess the micronutrient intake but there is a limited number of validated FFQs to assess the gluten intake¹⁵⁻¹⁷. Moreover, to the best of what is known, there is no information available on the amount of gluten consumed by the general population in Spain. Furthermore, information on gluten contained in commonly consumed foods is poor and difficult to obtain. Although in Spain there can be found some validated FFQs that collect information on different diet characteristics¹⁸, they do not include all the food products needed to assess gluten intake.

The purpose of this work is to develop and validate a FFQ in order to assess gluten intake (FFQ-gluten) in Spanish children aged 7 to 36 months in the area of Valencia (Spain) to be used in the Prevent CD study. Therefore, these specific FFQS could also prove useful for epidemiological studies, especially in the field of CD.

In addition, by applying this specific FFQ, the aim is to establish dietary habits in relation to gluten intake early in life in the population studied.

Subjects

The parents of children aged 7-36 months recruited in different nurseries and primary schools were asked to participate in the study. The selected children were attending primary healthcare centers for routine health surveys, they had minor health problems or minor surgery controlled at the Pediatric Department of the La Fe University Hospital in Valencia, and they were interviewed by the same dietitians' team.

The exclusion criteria were: (1) any oral-motor dysfunction, (2) diagnosed food allergy, (3) any gastrointestinal disease or any minor gastrointestinal disorder (diarrhea, constipation, gastroesophageal reflux) (4) malnutrition, and (5) children on dietary restrictions or following specific diets.

Methods

This study was conducted according to the guidelines set up in the Declaration of Helsinki and was approved by the Ethics Committee of the "Instituto de Investigación Sanitaria La Fe" (Valencia, Spain).

All parents were informed about the purpose of the survey and of the ultimate study aim. Informed participation consents were obtained from them all.

Development of FFQ

The development of a list of food products, including the item of interest for the study, is a crucial preliminary step to develop an FFQ. For a food item to be considered, it must be eaten reasonably often by a considerable number of people, and it must contain a substantial amount of the nutrient/food group of interest, which in this case study is gluten¹⁵. By national's laws, products with less than 20 mg/kg (200ppm) are considered as gluten free. Thus, a pilot survey was carried out among parents on their children's eating habits using household measures and 24-hour recalls. The pilot study started collecting prospectively information on the most frequent gluten-containing food, as well as brands and portion sizes, consumed by healthy infants aged between 7 and 36 months from Valencia. This lead to the selection of all the gluten-containing products to be included in the FFO template and the average portion size for each item; those food products containing only gluten traces were discarded. All these first tasks were performed by an expert nutritionist in the field.

The next step was to determine the amount of gluten in these products and then to gather the individual

items in food groups according to their origin and amount of gluten. Moreover, for products like bread, biscuits, infant cereals and pasta, the food groups were divided into subgroups as despite them having a similar composition, the amount of gluten differed significantly depending on the brand or the portion size consumed. For bread, all the identified brands and types were pooled under the same group, except for sliced bread typhus sandwich, which was considered separately because the portion size consumed was bigger than the white bread one. Ten brands of biscuits with different formulations were divided into three different subgroups because there were appreciable differences in gluten content among brands. All the infant cereal brands were included in the same subgroup, except for one because it had higher gluten content. Two different subgroups were established for pasta after considering that a different amount of the product is contained in a pasta dish as compared to soup with pasta.

The portion size of the individual groups was established according to the information provided by the preliminary survey: 1 slice of bread; 1 unit for biscuits and breadsticks; the number of spoonfuls for infant cereals; amount of raw pasta contained in the two different pasta meal types; individual portions in grams for the remaining products.

Moreover when the pilot survey was conducted, it was observed that after the age of 12 months, new and more complex gluten-containing foods were introduced. Thus, it was decided to develop two different food-frequency questionnaires (FFQs): FFQ-1 for infants aged 7 to 12 months (FFQ1-Online Resource-1) and FFQ-2 for children aged between 13 and 36 months (FFQ2-Online Resource-2). In the first one, 13 food groups were included and participants were also asked about age when the items were introduced; in the second one, 20 food groups were considered. In this group, however, the age at which the items were introduced was not asked. Complementary questions about breast-feeding, types and brands of formula feeding and infant cereals were added in both FFQs.

In both FFQs, frequencies of intake of the included food products were collected as follows: never, less than once a week, 1 to 3 times a week, 4 to 6 times a week, once a day, twice a day, and three or more times a day. For the once or twice a day frequency, participants were asked to specify the amount of food i.e. 3 biscuits. For three or more times a day, participants were asked to specify both the amount of food and the exact number of times per day their child ate this particular product. Finally, the exact number of spoonfuls was reported for infant cereals.

Assessing amount of gluten

To calculate the amount of gluten of all the items included in the FFQs, the method of Overbeek *et al.* was used. This method considers that gluten represents

about 80% of the total wheat proteins, and thus recommends multiplying the grams of gluten containing vegetable proteins of each product by 0.88,16,17. However, before this formula was applied, it was needed to establish the exact amount of gluten-containing vegetable proteins in each foodstuff and per brand. Therefore, as the information presented on food labels was usually inadequate -in most cases only the amount of total protein was indicated and no further specifications were shown- the data was obtained from different sources: WHO-FAO information from the Internet; companies' official websites; specific food composition tables; and, in many cases, directly contacting the company by either e-mail or telephone. Finally, a database of the amount of vegetable protein, the gluten content of all the different products and brands consumed by the infant population studied was created. It was considered gluten-containing food those which included wheat, barley, rye or oats. However, oats are very seldom consumed by infants and young children as part of the Mediterranean Spanish diet.

Conversion of frequency into mean daily gluten intake

The weekly frequency of intake of each product was converted into mean daily gluten intake (MDGI). First of all, the amount of gluten contained in an individual portion of all the food products was determined. This amount contributes to a "once a day" consumption, from which the rest of the frequencies were calculated according to the following approach:

- When products are eaten "Less than once a week", the amount of an individual portion was divided by 14 because we assumed that this frequency may be about once every 2 weeks. If parents specified that children ate, for example, 3 biscuits, but "less than once a week", the amount of 3 individual portions was divided by 14.

For the remaining frequencies, the following approach was adopted:

- The "1 to 3 times a week" frequency: the amount of an individual portion was divided by 7 days and multiplied by 2 (2 being considered the average of "1 to 3 times a week").
- The "4 to 6 times a week" frequency: the amount of an individual portion divided by 7 days and multiplied by 5 (5 being considered the average of "4 to 6 times a week").
- The "Once a day" frequency: the amount of an individual portion.
- The "Twice a day" frequency: the mount of an individual portion multiplied by 2.
- The "Three or more times a day" frequency: the amount of an individual multiplied by the number of times consumed.

The total MDGI in grams was obtained by adding them all up.

Questionnaire validation

To validate the FFQ, the study population was asked to fill in both the FFQ and a 2-day-record (2DR) consecutively by the same parents in two different months to be able to compare the results obtained from the two different surveys. The 2DR is a commonly accepted tool in food consumption studies. However in order to ensure FFQ reliability, the two FFQs were also compared against 7-day-record (7DR) in a smaller group of individuals. The sample size required to validate the FFQ against a 7DR depends on the differences in the mean gluten intake between the FFQ and 2DR and on the maximum accepted deviation between the results of the FFQ and the 2DR, which we established as $20\%^{8.9,12,19,20}$.

Firstly, the participants completed the FFQ and, the next month, a 2DR (1 weekday and 1weekend day from the same week). Afterwards the difference of the mean obtained was calculated by each method and, according to this difference, it was possible to calculate the number of participants who had to fill the FFQ and 7DR in the same way that FFQ and 2DR.

Statistical Analysis

Statistical analysis and charts were performed with the R software (version 3.0.2). Data were described with the mean and standard deviation. The sample was formed by 342 children (126 were aged 7-12 months and 216 were aged 13-36 months). The Bland-Altman plots stratified by age group were used to assess the agreement between 2DR vs FFQ and 7DR vs FFQ methods. Additionally, average bias and the 95% limits of agreement were computed for each agreement plot. As an alternative agreement measure, Lin's concordance correlation coefficient was also computed in each case²¹.

Results

Study population

The study population was formed by 342 children; within this amount, 126 were aged 7-12 months (73 girls and 53 boys), they were pooled as group 1 and they contributed to FFQ1 development; 216 were aged 13-36 months (123 girls and 93 boys), i.e. group 2, and contributed to FFQ2. The profile of the majority of the parents who participated was: non-smokers, primary educational level, married, with more than one child, Caucasian, without social problems and some of them did some physical activity at least once a week.

All of them were native from the Regional Community of Valencia, a geographical area with around 5 million inhabitants, coming from different urban and rural settings.

In group 1, the MDGI obtained by the 2DR and the FFQs was 2.81g/day and 2.67 g/day, respectively. According to the difference between these means, it was statistically determined that in order to validate FFQ1, at least 15 parents had to complete the FFQ and the 7DR; the MDGI obtained by 7DR and FFQ was 2.44 g/day and 2.47 g/day respectively.

In group 2, the MDGI obtained by 2DR and the FFQs was 4.85 g/day and 5.68 g/day respectively. According to the difference between these means, it was statistically determined that to validate FFQ2, at least 30 parents had to complete the FFQ and the 7DR. The MDGI obtained by 7DR and FFQ was 4.4 g/day and 4.15 g/day respectively. The mean gluten intakes obtained by the FFQ, the 2DR and 7DR are shown in table I.

Different sources of gluten

The data obtained from the two FFQs allowed to establish the percentages that each food group contributed to the MDGI. The population was divided into different age groups to show the evolution of gluten-containing product intake.

Table I
Mean daily gluten intake in grams per day and the standard deviation (SD)
obtained by 2DR, 7DR and FFQ in both age groups

	Age (months)	n	2DR MDGI (SD)	7DR MDGI (SD)	FFQ MDGI (SD)
Study 1	7-12	126	2.81 (1.66)		2.67 (1.11)
(2DR vs FFQ)	13-36	216	4.85 (2.19)		5.68 (1.99)
Study 2	7-12	15		2.44 (0.66)	2.47 (1.58)
(7DR vs FFQ)	13-36	30		4.40 (1.58)	4.15 (1.35)

2DR: 2-day food record; 7DR: 7-day food record; MDGI: Mean daily gluten intake in grams per day; SD: Standard Deviation; FFQ: food frequency questionnaire.

 Table II

 Contribution of different gluten sources as a percentage (%) of total gluten intake at different age points.

	7-8 months	9-10 months	11-12 months	13-18 months	19-24 months	25-36 months
% of food products	8					
Infant's cereals	60,67	53,86	52,19	24,34	18,26	13,01
Bread	24,86	23,4	16,54	28,0	35,40	40,21
Biscuits	14,46	13,64	11,9	16,45	14,8	12,63
Pasta	0,00	0,25	1,20	3,40	6,40	8,70
Others	0,00	8,85	18,17	27,8	25,13	25,45

As displayed in table II, intake of bread, pasta and other food products (baby food, crumbed products) progressively increased with age at the expense of eating less infant cereals.

Statistical results

Average bias comparing 2DR and FFQ was -0.14 g/day in the 7-12 age group (95% CI [-0.30, 0.03]) and 0.40 in the 13-36 group (95% CI [0.08, 0.71]). 95% limits of agreement ranged from -1.95 to 1.68 in the 7-12 months age group and from -4.23 to 5.03 in the 13-36 months group. There was a noticeable increase in the limits of agreement in the higher age group (Fig. 1). Results of Lin's concordance correlation coefficient were consistent with those obtained from Bland-Altman plot with $\rho=0.76$ (CI 95% [0.67, 0.82]) for the 7-12 age group and $\rho=0.57$ (CI 95% [0.48, 0.65]) for the 13-36 age group, representing an acceptable correlation between 2DR and FFQs in both age groups. When comparing 7DR and FFQ, average bias was 0.03 g/day in the 7-12 age group (95% CI

[-0.20, 0.27]) and -0.25 in the 13-36 group (95% CI [-0.94, 0.44]) with 95% limits of agreement ranging from -0.8 to 0.87 in the 7-12 age group and from -3.87 to 3.34 in the 13-36 age group. As in 2DR validation, the limits of agreement increased in the high age group (Fig. 2). As expected, the Bland-Altman plot with limits of agreement between 7DR and FFQ provided better results, with observations coming close to zero for both age groups, which reflects the similarity of the results when either instrument is used. Lin's concordance correlation coefficient was ρ =0.77 (95% CI [0.45, 0.91]) for the 7-12 age group and ρ =0.61 (CI 95% [-0.15, 0.52]) for the 13-36 age group, being in both age groups an acceptable correlation.

Discussion

For the first time, it has been developed a tool to assess gluten intake in infants aged 7-36 months from Valencia. Although the Spanish dietary habits vary throughout the different geographical regions, the same gluten containing products (although in different

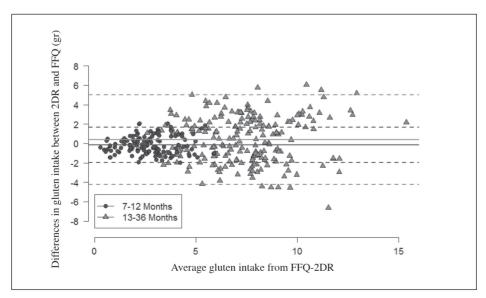


Fig. 1.—Bland-Altman plot of the gluten intake according to the FFQ and the 2DR for the studied sample.

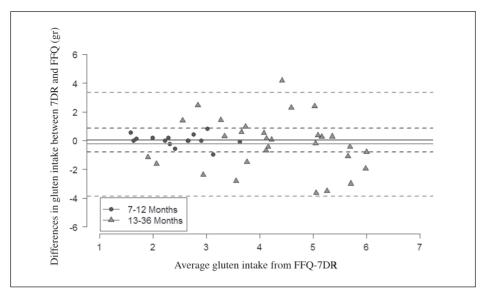


Fig. 2.—Bland-Altman plot of the gluten intake according to the FFQ and the 7DR for the studied sample.

proportions) are consumed all over the country. Therefore it can be assumed these FFQs could be used not only in Valencia but also for the general Spanish population aged 7 -36 months. No prevailing food record instrument was used, but two completely new FFQs were developed after conducting a preliminary survey on dietary habits of an age-matched population.

Validation of an FFQ method prior to being used is essential to avoid false associations between dietary factors and diseases, or disease-related markers. Thus validation studies have to be carried out to assess whether the questionnaire measures correctly or to assess if it is in line with an accepted "gold standard" 19,22,23. A 7DR is considered the gold standard for diet-based studies as it allows the collection of products consumed a few times a week, or even only once. Hence the information included is more accurate than that obtained by a 2DR. Nevertheless, the 2DR usually provides enough information and is easier for parents to carry out^{8,22,24}. Comparing first the results obtained by the FFQ and the 2DR enables to determine the exact number of 7DR needed for validation, thus minimizing the amount of work done by the collaborating parents. As shown, two FFQs were successfully validated against both 2DR and 7DR. Very few differences were found between the data obtained by the FFQs-gluten and the 7DR, thus indicating that the FFQs-gluten contemplates all the gluten-containing products usually consumed by the infant population, and that the developed gluten-FFQs used, proves a better tool than 2DR to determine the MDGI for infants aged 7-36 months. FFQ is superior to 2DR, especially in infants older than 13 months of age as wider food diversity leads to some products not being eaten on a daily basis, which can be easily missed by short food surveys.

Furthermore, these two FFQs allowed to establish for the first time gluten intake habits in Spanish infants. Between the ages of 7 and 12 months, only a limited diversity of gluten-containing food is introduced. Yet the

older the study population becomes, more new products are introduced. Consequently, the dispersion between the two methods, 2DR-FFQ and 7DR-FFQ, is greater for the 13-36 months age group as compared to the younger group. This diversity also accounts for a higher MDGI in the 13-36-month-old infants, which is almost twice the amount recorded for the younger age group. The amount of gluten provided by cereal intake progressively decreases and contributed to about 60% of gluten intake at the age 7 months, but to only 14% at 36 months of age. Conversely, bread and pasta intake increases gradually together with other products, such as breadsticks (similar to bagel), coated foods (in breadcrumbs or flour), etc.

The two FFOs developed are crucial for precise gluten intaking quantification in specific surveys, such as the Prevent CD study whose results have recently been published (10). In this prospective double blind study it has been shown that the introduction of small quantities of gluten at 16 to 24 weeks of age did not reduce the risk of CD at 3 years of age in a group of high-risk children; it was also determined that gluten intake up to 36 months of age was not related to the development of CD. These associations could be studied thanks to the application of these and other similar FFQs specifically developed for this purpose. Moreover, in the latest study by Anneli Ivarsson and co-workers, an increase was observed in the CD incidence from 1973 to 2009, although the infant feeding recommendation had not been changed since 1995. The group suggested that other environmental and lifestyle factors, such as an increase in gluten consumption, may explain the observed changes in the celiac disease epidemic²⁵. In any case, further impact of gluten intake in CD development later in life is still to be determined.

Very few studies on gluten intake in different populations have been published up to date. A survey in Dutch population using a 2DR showed gluten consumption was 12.0 g/day, which is twice that obtain

ned for the older age group studied. However, included subjects were aged 2-57 years and no information about MDGI for different age ranges was provided²⁰. Therefore, a comparison with the data is not feasible.

So far, no other specific FFQ to assess gluten intake has been published in other countries, except for the Netherlands, where the authors developed and validated first an FFO for 0-12-month-old infants and more recently one for children aged 1 to 4 years; in both cases a MDGI about twice that recorded in this study for the same age ranges was reported8,9. The gluten intake pattern of Dutch children is, however, similar to that of the Spanish infants as far as a decrease in porridge intake and an increase in bread intake in relation to age was observed; as for biscuits, no relevant variation was found for the observed age range for both the Dutch and Spanish populations. Nevertheless, many food items included in the Dutch FFQ are only seldom, if not at all, consumed by Spanish children in the geographic area analyzed; conversely, some food habits in Spain include products that are not available in other countries, which also mean that the Dutch FFQ cannot be applied in the setting.

Implications for Research and Practice and Conclusion

The specific FFQs-gluten developed are the only validated questionnaires available to determine the amount of gluten consumed in the pediatric age in the geographical region analyzed. They are user-friendly and are excellent instruments to assess gluten intake in children aged 7-36 months, as well as allowing comparisons of gluten intake to be made in different populations.

Reliable and precise information on gluten intake habits is crucial to conduct epidemiological studies in the field of CD and to establish the real association between gluten consumption and development of CD. For such purposes specifics tools as these easy-to-use FFQs are necessary.

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FFQ1. Food frequency questionnaire for Spanish children at 7 – 12 months of age.

Name (Initials): Date of birth of your child: Sex: Date of filling the questionnaire:	Food Questionnaire
1. Does the child exclusively breastfeeding?	☐ No ☐ Yes, go to question number 4
3. What kind of feeding formula do you give your child?	☐ Normal Amount: Commercial brand:
	Adapted to food allergy Amount: Commercial brand:
	Antireflux / antiregurgitation Amount:
	Others Amount: Commercial brand:
4. Do you feed your child with gluten containing cereals?	☐ No, go to question number 6.☐ Yes. Commercial brand:
5. How much cereal do you feed your child per day? Indicate the number of scoops or spoonfuls, either in bottle or porridge.	☐ 1 - 3 scoops / spoonful: ☐ Once a day ☐ 2-3 times a day ☐ 4-5 times a day ☐ 4-6 scoops / spoonful: ☐ Once a day ☐ 2-3 times a day ☐ 4-5 times a day ☐ 4-5 times a day ☐ 4-5 times a day ☐ G-7 times a day ☐ G-7 times a day ☐ Once a day ☐ 2-3 times a day ☐ 1 Once a day ☐ 2-3 times a day ☐ 2-1 times a day ☐ 2-3 times a day ☐ 3-1 times a day ☐ 4-5 times a day ☐ 4-7 times a day ☐ 6-7 times a day
	much cereals and what commercial brand do you feed your child?
6. When you give salted jars (not homemade) to your child, which commercial brand do you use?	☐ I don't use "ready-to-feed meals" ☐ Hero: - Good night (vegetables and pasta cream) - Market (vegetables with turkey and pasta) ☐ Others:
7. When you give fruit jars to your child, which commercial brand do you use?	☐ I don't use "ready-to-feed meals"☐ Hero: fruit and cereals macedoine☐ Nestlé: fruit and cereals cocktail☐ Others:
8. With what frequency do you give your child the follo	wing food products?

FFQ1 (continued).

		FRE	FREQUENCY	λũ							A.	GE O	AGE OF INTRODUCTION	SODU	CTION				
FOOD PRODUCTS	NEVER	Less than once a week	1-3 times a week	4-6 times a week	Once a day	Twice a day	3 times or more a day	м9>	6-7 m	<7m /	7-8 m s-7		8-9 m	5 m6>	<9m 9-10m <10m	<10m	10- 11m	<11m	11- 12m
Bread (1 slice = $40 g$)																			
Biscuits (María, María Dorada, Rio Petit y Tosta Rica)																			
Dinosaurios Lú biscuits																			
Cake $(1 \text{ portion} = 20g)$																			
Pastry (1 portion =35g)																			
Breadsticks (Unit)																			
Chocolate (1 unit)																			
Cereals (Nestlé, Milupa, Ordesa, Hero Baby)																			
Cereals (Nutribén)																			
Pasta: (stars, wheat semolina, letters, etc) (Small dish)																			
Crumbed products $(1 \text{ portion} = 75g)$																			
Fruit jars (with gluten)																			
Salted jars (with gluten)																			
*In the ontion "3 times or more a day" specify the amount	timee or m	ore a da	TO CHOC	ifty the ar	nount														

*In the option "3 times or more a day", specify the amount.

FFQ2. Food frequency questionnaire for Spanish children at 13 – 36 months of age.

Name (Initials): Date of birth of your child: Sex: Date of filling the questionnaire:	Food Questionnaire
1. Does the child exclusively breastfeeding?	☐ No ☐ Yes, go to question number 4
3. What kind of feeding formula do you give your child?	Normal Amount: Commercial brand:
	Adapted to food allergy Amount: Commercial brand:
	Antireflux / antiregurgitation Amount:
	Others Amount: Commercial brand:
4. Do you feed your child with gluten containing cereals?	☐ No, go to question number 6.☐ Yes. Commercial brand:
5. How much cereal do you feed your child per day? Indicate the number of scoops or spoonfuls, either in bottle or porridge.	☐ 1 - 3 scoops / spoonful: ☐ Once a day ☐ 2-3 times a day ☐ 4-5 times a day ☐ 4-6 scoops / spoonful: ☐ Once a day ☐ 2-3 times a day ☐ 4-5 times a day ☐ 6-7 times a day ☐ Once a day ☐ 2-3 times a day ☐ 2-3 times a day ☐ 4-5 times a day ☐ 2-7 times a day ☐ 4-5 times a day ☐ 4-7 times a day
	your child?
6. When you give salted jars (not homemade) to your child, which commercial brand do you use?	☐ I don't use "ready-to-feed meals" ☐ Hero: - Good night (vegetables and pasta cream) - Market (vegetables with turkey and pasta) ☐ Others:
7. When you give fruit jars to your child, which commercial brand do you use?	☐ I don't use "ready-to-feed meals" ☐ Hero: fruit and cereals macedoine ☐ Nestlé: fruit and cereals cocktail ☐ Others:
8. With what frequency do you give your child the follo	wing food products?

FFQ2 (continued).

		FREC	FREQUENCY				
FOOD PRODUCTS	NEVER	Less than once a week	1-3 times a week	4-6 times a week	Once a day	Twice a day	3 times or more a day
Bread (1 slice = $40 g$)							
Sandwich bread (1 slice = 30g)							
Biscuits (María, María Dorada, Rio Petit y Tosta Rica)							
Dinousaurius Lu biscuits							
Chocolate biscuits (such as Principe)							
Cake $(1 \text{ portion} = 20g)$							
Pastry (1 portion = 35g)							
Breadsticks (Unit)							
Cereals (Nestlé, Milupa, Ordesa, Hero Baby)							
Cereals (Nutribén)							
Breakfast cereals (Kellogs)							
Chocolate cereals							
Pasta: (stars, wheat semolina, letters, etc) (Ration of soup)							
Pasta: (spaghetti, macaroni, ties, etc) (Small dish)							
Hake sticks (1 portion = $75g$)							
Chicken nuggets (1 portion = 75g)							
Crumbed products (meat, fish, cheese, etc) (1 portion =75g)							
Milk products with flour (cereals or biscuits)							
Fruit jars (with gluten)							
Salted jars (with gluten)							

*In the option "3 times or more a day", specify the amount.