

Nutritional status and dietary factors in cystic fibrosis patients with delta F508 mutation

Estado nutricional e fatores dietéticos de pacientes com fibrose cística portadores da mutação delta F508

Carina de Sousa SANTOS¹
Thais STEEMBURGO²

ABSTRACT

Objective

To examine the association between nutritional status and dietary factors in children and adolescents with cystic fibrosis that are carriers of delta F508 mutation.

Methods

Cross-sectional study of cystic fibrosis children and adolescents. Nutritional status (body mass index percentile) and dietary intake (3-day diet records presented as a percentage of estimated energy requirement) were assessed.

Results

Thirty six patients (median of 8.6; interquartile range 6.8-12.5 years; 50% male). The Poisson regression analysis showed that the carriers for delta F508 mutation had 60% lower prevalence *ratio* of body mass index $\geq 25^{\circ}$ ($PR=0.4$; 95% $IC=0.2-0.8$) and 90% lower prevalence *ratio* ($PR=0.1$; confidence interval 95% $IC=0.02-0.3$) of $\geq 150\%$ of estimated energy requirement when compared with non-delta F508 mutation carriers. The model was adjusted for lung function, estimated energy requirement, and body mass index.

¹ Universidade Federal de Santa Catarina, Centro de Ciências da Saúde, Programa de Pós-Graduação em Nutrição. Florianópolis, SC, Brasil.

² Universidade Federal do Rio Grande do Sul, Centro de Estudos em Alimentação e Nutrição, Departamento de Nutrição. R. Ramiro Barcelos, 2400, Rio Branco, 90035-003, Porto Alegre, RS, Brasil. *Correspondência para/correspondence to:* T STEEMBURGO. E-mail: <tsteemburgo@gmail.com>.

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Conclusion

Carriers for delta F508 mutation showed lower body mass index percentile and lower daily caloric consumption when compared with patients without this mutation.

Keywords: Cystic fibrosis. Diet. Nutritional status.

RESUMO

Objetivo

Avaliar a associação entre estado nutricional e fatores dietéticos na fibrose cística em crianças e adolescentes portadores da mutação delta F508.

Métodos

Estudo transversal com crianças e adolescentes com fibrose cística. Foram avaliados o estado nutricional (índice de massa corporal em percentil) e consumo alimentar (três registros alimentares apresentados em porcentagem da necessidade de energia estimada).

Resultados

Foi avaliado um total de 36 pacientes (mediana de 8,6; intervalo interquartil de 6,8-12,5 anos; 50% do sexo masculino). Na regressão de Poisson, pacientes com a mutação delta F508 apresentaram uma razão de prevalência 60% menor de índice de massa corporal $\geq 25^{\circ}$ (RP=0,4; IC95%=0,2-0,8) e uma razão de prevalência 90% menor de $\geq 150\%$ da necessidade de energia estimada (RP=0,1; IC95%=0,02-0,3) quando comparados àqueles com ausência dessa mutação. Modelo ajustado para função pulmonar, necessidade de energia estimada e índice de massa corporal.

Conclusão

Pacientes portadores da mutação delta F508 apresentaram um menor percentil de índice de massa corporal e menor consumo calórico diário quando comparados àqueles sem a mutação.

Palavras-chave: Fibrose cística. Dieta. Estado nutricional.

INTRODUCTION

Cystic fibrosis is characterized by diverse clinical expressions and a considerable variability intensity and rate of progression¹. The heterogeneity of such phenotypes may be attributed to 1,940 mutations already identified in the Cystic Fibrosis Transmembrane Conductance Regulator (CFTR)², which are classified as class I to V according to the mechanisms through which they disrupt the membrane protein function³.

There is a substantial interest in the influence of the genotype on the cystic fibrosis phenotype, but the use of class of mutations to establish associations should be intended for population studies only and not for individual prediction purposes because they are not designed to serve as an instrument for clinical prognosis⁴. Therefore, studies on the genotype

and phenotype relationships focus on the investigation of delta F508 mutation (deletion of *Phenylalanine* at 508)⁴, which is the most frequent mutation, found in approximately 70% of alleles worldwide².

Delta F508 homozygous individuals often have more pronounced clinical manifestations compared to other genotypes: they have higher levels of sweat chloride⁵, are more susceptible to pancreatic insufficiency^{5,6}, and have more severe lung insufficiency, leading to an early diagnosis of the disease^{5,6} and higher mortality risk⁵. Although there are studies addressing the relation between delta F508 mutation and nutrition-related phenotypes in cystic fibrosis, such as poor nutritional status⁵⁻⁸, high energy consumption^{9,10}, presence of *Mellitus* diabetes¹¹, and decreased levels of essential fatty acids¹², these factors have not been entirely elucidated yet.

It is known that the nutritional conditions of cystic fibrosis individuals directly affect the clinical course of the disease, and irrespective of the genotype, an increase by 10 to 100% in Estimated Energy Requirement (EER) is recommended to achieve optimum weight gain¹³.

In this regard, knowing that the type of mutation present in the CFTR gene of cystic fibrosis may determine the between nutritional status and dietary factors in cystic fibrosis children and adolescents that are carriers for delta F508 mutation.

METHODS

This is a cross-sectional study on cystic fibrosis children and adolescents that are under treatment at the Joana Gusmão Children Hospital, a reference center in the state of *Santa Catarina* (*Florianópolis*, Brazil) for pediatric cystic fibrosis treatment. Children aged ≥ 5 years (age at which pulmonary function tests starts) who were tested for the presence of delta F508 mutation and did not have any associated disease that could affect the outcome of the study were invited to participate in the research. Data was collected once a week from September/2012 to June/2013 on the day of the routine treatment of cystic fibrosis at the hospital. The invitations to participate were sent out by the researcher in charge, and the study was carried out after the participants signed the Free and Informed Consent Form.

Participants were divided into three groups according to the presence of mutation: homozygous for delta F508, heterozygous for delta F508, and non-delta F508 mutation. The genotyping test consisted of a screening of the most frequent mutations using fluorescent allele-specific polymerase chain reaction (TaqMan® SNP Genotyping Assays; Applied Biosystems, Foster City, California, United States).

The variables were: gender (female/male); patient age (years), age and education (years) of

patient parent or guardian and their economic level, which was estimated based on their purchasing power¹⁴ (class B/class C - there were no families belonging to classes A, D, and E).

The patient's medical record variables were: use of pancreatic enzymes (yes/no); use of antibiotics and pulmonary bacterial colonization during the period of data collection (yes/no); and type of pulmonary bacterial colonization (*Burkholderiacepacia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and methicillin-resistant *Staphylococcus aureus*). The colonization-type test was performed by collecting material from the oropharynx. Bacteriological determination was performed using the quantitative method, and a count of $\geq 10^4$ CFU/mL indicated presence of infection^{15,16}.

Lung function was assessed by the estimated value of the Forced Expiratory Volume in the first second (FEV₁) as percentage and prevalence *ratio* of <70 and $\geq 70\%$ using a spirometer (Spirolab®, Medical International, Research, Rome, Italy). The 70% cutoff value, considered as mild obstruction, is the last cutoff value for this classification¹⁷.

Variables for assessment of the nutritional status

To assess the patients' nutritional status, Triceps Skinfold (TSF), Mid-Arm Muscle Circumference (MAMC) and Body Mass Index (BMI), which are indicators recommended by the American Consensus of Nutrition for cystic fibrosis¹⁸, were used.

To calculate Mid-Arm Muscle Circumference (centimeter [cm]), the Mid-Arm Circumference (MAC) (cm) and TSF (millimeter [mm]) were measured following protocols¹⁹. MAC was measured using a Cescorf® steel tape, 0.10 mm sensitivity in 1 mm (*Cescorf Equipamentos para Esporte Ltda*®, *Porto Alegre* [RS], Brazil). TSF was measured using a Lange Skinfold Caliper® (Beta

Tecnology Inc., Cambridge, Maryland, United States) 0-60 mm scale, 1 mm accuracy, and constant pressure of 10 g/mm² at any aperture. Using MAC and TSF, MAMC was calculated according to the following formula: ($= \text{MAC} - 3.1416 \times [\text{TSF}/10]$)¹⁹. MAMC and TSF were also presented as a percentage of adequacy to fifth percentile, according to the following formula: [$= \text{resulting MAMC/TSF} / (\text{MAMC/TSF } 5^{\text{th}} \text{ percentile}) \times 100$]. The percentile values were related to the values of the National Health and Nutrition Examination Survey¹⁹.

To calculate body mass index, the weight (kilogram [kg]) and height (meters [m]) were measured following protocols²⁰. The weight was measured using a BK50F digital scale with 150 kg maximum capacity and 0.1 kg resolution (Balmak®, Santa Barbara d'Oeste [SP], Brazil). Height was measured using an anthropometer, with precision of 0.1 cm (Alturaexata®, Belo Horizonte, Brazil). BMI ($= \text{weight}/\text{height}^2$)²⁰ was calculated (percentile) based on the weight and height using the Stata® software, v.11.0 (Stata Corp, College Station, Texas, United States). The growth charts used were those developed by the World Health Organization²¹. BMI was expressed as percentile and prevalence of <25° and ≥25°. Cutoff was defined according to American consensus on nutrition for cystic fibrosis, which considers BMI ≥25° as acceptable¹⁸.

Variables for dietary intake assessment

Dietary intake data collection

Dietary intake assessment was based on 3 non-consecutive-day diet records (two week days and one day on the weekend) completed at home by the parents. On the day of data collection the patients' parents or guardians were given oral and written instructions on how to complete the diet records. The data (3 reports) was collected over the phone at a time convenient to the respondents.

Assessment of dietary intake

Calories, macronutrients, and dietary fibers intake were calculated using the Avanutri® software v.4.0 (Avanutri & Nutrição Serviços, Três Rios [RJ], Brazil), based on the Brazilian Table of Foods Composition²², and whenever the food was not described in the table used as reference, the product's label was used. In order to determine the average intake from the 3-day diet records, two methods of adjustment were used. The first adjustment was the within-person and between-person variance, which resulted in a corrected distribution using an error correction model with formula syntax in Excel® 2010 (Microsoft Corporation)²³. The second adjustment was made for total energy intake using the residual method and the linear regression model with the Stata® software, version 11.0. The consumption of each macronutrient was considered as a dependent variable, and the total caloric value as an independent variable²⁴. The total average energy intake over the three days after adjustment was compared to the EER of the Dietary Reference Intakes²⁵.

Presentation of variables to assess dietary intake

Energy intake was expressed as: Total Caloric Value (TCV), considering the daily amount of calories and as a percentage higher than EER (% EER).

Macronutrients and dietary fibers were expressed in grams/day. The macronutrients were also expressed as a percentage in relation to the TCV values (% TCV).

Total caloric value was also shown in three categories of percentages higher than EER (% EER) <110% EER; ≥110% EER - <150% EER and ≥150% EER. A cutoff value of 10% was chosen based on a systematic review as requested by the Cystic Fibrosis Foundation, in which, due to the caloric imbalance that occurs in such individuals, some authors¹³ recommended an additional

intake from 10 to 100% of the total daily caloric value suggested for healthy individuals. The percentage of contribution of the macronutrients was also investigated according to the minimum and maximum average intake of each macronutrient of the sample.

Quality control and statistical analysis

Data entry was performed with double data entry using the EpiData® software, version 3.1 (The EpiData Association, Odense, Denmark), followed by the external consistency test. Subsequently, and the data were analyzed using the statistical Stata® software, version 11.0.

With regard to the categorical outcome, the Chi-square test of heterogeneity was used. This test was replaced with the Fisher test when there were estimated values <5 in more than 20% of the cells. When there was a symmetrical numerical outcome, the Analysis of Variance (Anova) test (heterogeneity and trend) was used, and when it was asymmetrical, the Kruskal-Wallis test was used. Categorical variables were described as prevalence at a 95% confidence interval. Symmetrical numeric variables were expressed as mean and standard deviation (\pm), and asymmetrical variables as median and Interquartile (IQ) range.

Poisson regression analysis was performed with a robust error variation to construct the adjusted model, and the variables were categorized as follows: exposure: presence and absence of delta F508 mutation and outcomes of nutritional status: BMI <25° and \geq 25° and dietary intake: <150% EER and \geq 150% EER.

In this model, possible confounding variables were adjusted forward, and the variables that showed a p -value \leq 0.25 in the Wald test, when associated with the exposure and outcome, were considered; the variables with p -value <0.05 remained in the final model. The p -value of 5% (p <0.05) was considered the statistical significance level in all analyses performed.

The present study was approved by The Research Ethics Committee for Research Involving Humans of the *Joana Gusmão* Children Hospital (Register nº 011/2012, and opinion issued by the National Research Ethics Commission under nº 03/2012).

RESULTS

The sample comprised 36 patients (median of 8.6; IQ=6.8-12.5 years; 50.0% male), among which 27.7% (n=10) were homozygous for delta F508, 47.3% (n=17) heterozygous for delta F508, and 25.0% (n=9) did not have this mutation. Figure 1 shows the steps for the sample selection.

The socio-demographic and clinical characteristics are described in Table 1. The patients' age (9.7; IQ=8.0-12.9 years; p =0.030) and the parents or guardians' age (42.8 ± 2.6 years; p =0.014) were higher for the patients who were homozygous for delta F508; both results showed significant difference. No significant difference was observed in the parents or guardians' educational and economic level groups. Although the prevalence of bacterial colonization was higher between the patients homozygous for delta F508 (80.0%; 5%CI=61.9-93.7; p =0.287), this difference was not significant in the comparison between the groups and for the other clinical characteristics.

Indicators of the patients or guardians' nutritional status are shown in Table 2. Patients carrying delta F508 homozygotes had lower BMI percentile (21.9 ± 23.3 ; p =0.017) when compared to the other groups. A weak relationship was observed when the prevalence of BMI \geq 25 was assessed, and the patients homozygous for delta F508 had a lower value (40.0%; 5%CI=18.1-61.5; p =0.077) than those of the other two groups. However, when the prevalence of BMI \geq 25° was assessed between the patients homozygous and heterozygous for delta F508 and those with absence of F508 mutation, a significant statistical association between these two groups was observed (48.1% presence *versus* 88.9%

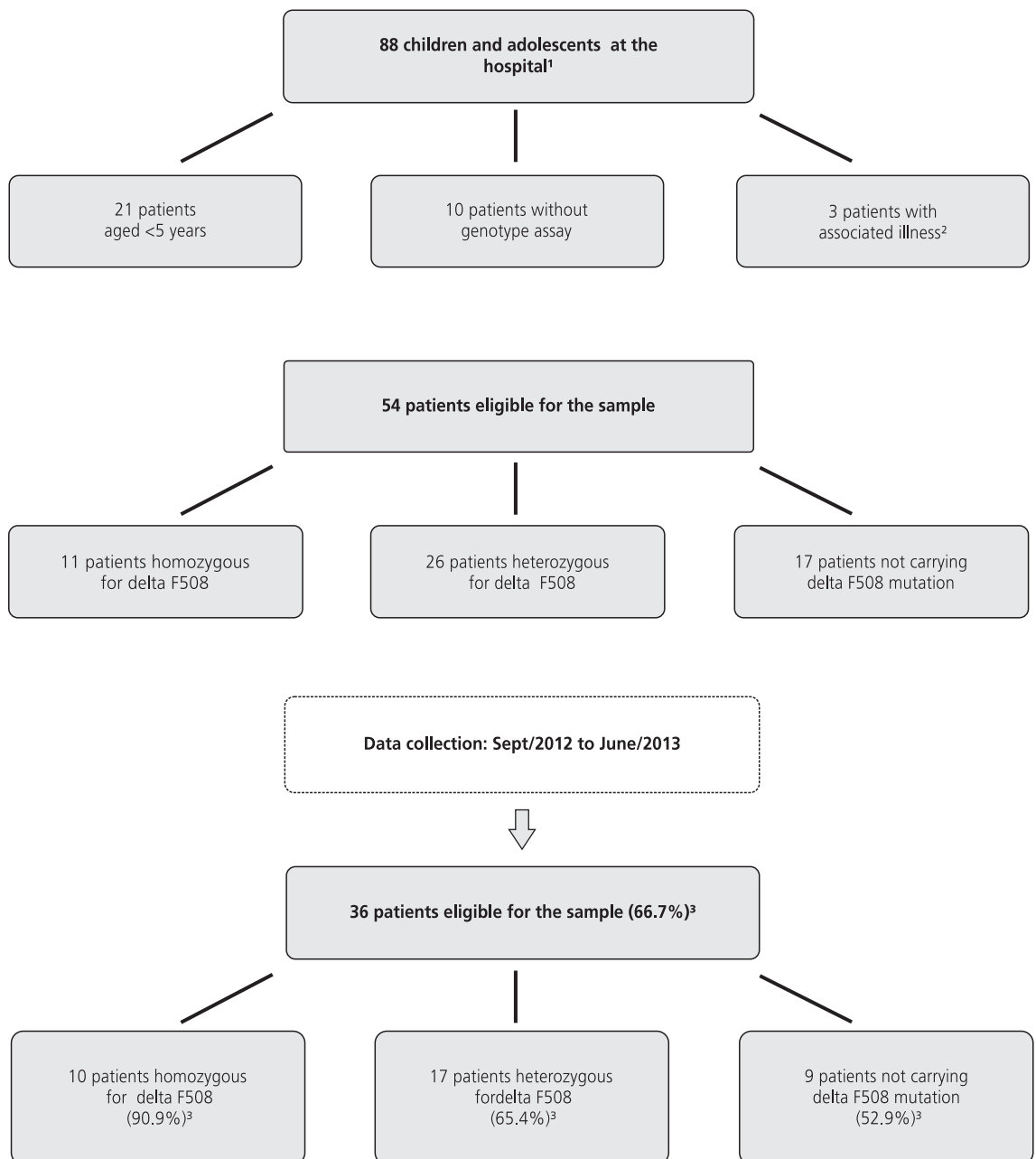


Figure 1. Flowchart of the steps for sample selection. *Florianópolis* (SC), Brazil, 2012-2013.

Note: ¹Number of children and adolescents with cystic fibrosis admitted to the *Joana Gusmão* Children Hospital in 2013, a reference center in the state of *Santa Catarina* (*Florianópolis*, Brazil) for pediatric treatment of cystic fibrosis; ²Microcephaly, congenital hypothyroidism, and dwarfism;

³Percentage of the total (n=54) number of eligible patients and total number of patients in each eligible group.

absence, Chi-square test $p=0.032$) (data not included in the Table). No statistical difference was found for the other nutritional status indicators.

The consumption of calories, macronutrients, and dietary fibers is shown in Table 3. When the

percentage of TCV was higher than the EER was assessed, the patients with delta F508 mutation, either homozygous ($130.6 \pm 17.8\%$) or heterozygous ($115.0 \pm 32.5\%$), showed lower consumption compared to the non-delta F508 patients

Table 1. Socio-demographic and clinical characteristics according to the presence of delta F508 mutation. Florianópolis (SC), Brazil, 2012-2013.

Characteristics	Total n=36		Non-delta F508 carriers n=9		Heterozygous for delta F508 n=17		Homozygous for delta F508 n=10		p-value
<i>Gender¹</i>									
Male	50.0	(26.0-74.0)	55.6	(30.7-78.5)	47.1	(21.5-69.2)	50.0	(26.0-74.0)	0.919*
Female	50.0	(26.0-74.0)	44.4	(21.5-69.2)	52.9	(30.7-78.4)	50.0	(26.0-74.0)	
<i>Age (years)²</i>									
Age (years) ²	8.6	(6.8-12.5)	7.3	(6.3-7.6)	9.1	(7.2-12.5)	9.7	(8.0-12.9)	0.030**
<i>Parents or guardians' age (years)³</i>									
Parents or guardians' age (years) ³	38.6	±7.2	33.4	±1.3	38.7	±1.6	42.8	±2.6	0.014†
<i>Parents or guardians' educational level (years)²</i>									
Parents or guardians' educational level (years) ²	8.5	(4.5-12)	7.0	(5-11)	9	(7-12)	7.5	(4-15)	0.762**
<i>Socio-economic level¹</i>									
Class B (B1 and B2)	50.0	(26.0-74.0)	55.6	(30.7-78.5)	52.9	(30.7-78.4)	40.0	(17.3-64.2)	0.752*
Class C (C1 and C2)	50.0	(26.0-74.0)	44.4	(21.5-69.2)	47.1	(21.5-69.2)	60.0	(35.7-82.7)	
Use of pancreatic enzymes ¹	91.6	(75.6-98.1)	88.9	(71.8-96.6)	88.2	(71.8-96.5)	100.0	(0.0-15.7)	0.866**
Use of antibiotics ¹	58.3	(34.0-78.2)	77.8	(52.8-91.8)	47.1	(25.7-70.2)	60.0	(38.4-81.9)	0.350**
<i>Pulmonary bacterial colonization¹</i>									
<i>Burkholderiacepacia</i>	7.4	(0.0-84.2)	20.0	(0.0-84.2)	0.0	0.0	12.5	(0.0-84.2)	0.938**
<i>Pseudomonasaeruginosa</i>	29.6	(3.2-65.0)	20.0	(3.2-65.0)	35.7	(8.5-75.5)	25.0	(3.2-65.0)	
<i>Staphylococcus aureus</i>	25.9	(0.3-70.9)	20.0	(0.3-57.9)	28.5	(0.3-70.9)	25.0	(0.3-70.9)	
Methicillin-resistant <i>Staphylococcus aureus</i>	37.0	(12.1-73.7)	40.0	(12.1-73.7)	35.7	(12.1-73.7)	37.5	(12.1-73.7)	
<i>Lung Function¹</i>									
<70% FEV ₁	33.3	(9.9-65.1)	22.2	(5.4-57.2)	47.1	(21.1-78.9)	20.0	(2.0-48.4)	0.346**
≥70% FEV ₁	66.7	(44.7-84.4)	77.8	(57.8-92.8)	52.9	(32.8-74.4)	80.0	(57.8-92.8)	
Lung Function (FEV ₁) ²	81.8	(52.7-95.0)	90.7	(74.1-95.2)	74.6	(42.5-87.5)	89.2	(76.8-109.7)	0.236**

Nota: Values expressed as: ¹Prevalence and confidence interval of 95%; ²Median and Interquartile range (25-75%); ³Mean and standard deviation. *Chi-square test of heterogeneity; **Fisher's exact test; †Analysis of Variance test of heterogeneity; **Kruskal-Wallis test. FEV₁: Forced Expiratory Volume in the first second.

Table 2. Nutritional status according to the presence of delta F508 mutation. Florianópolis (SC), Brazil, 2012-2013.

Variables	Total n=36		Non-delta F508 carriers n=9		Heterozygous for delta F508 n=17		Homozygous for delta F508 n=10		p-value
BMI (percentile) ¹	33.0	±27.9	52.2	±29.3	30.1	±26.1	21.9	±23.3	0.017**
BMI ≥25° (%) ²	58.3	(34.0-78.2)	88.9	(69.6-98.8)	52.9	(29.8-74.3)	40.0	(18.1-61.5)	0.077**
TSF (mm) ³	10.9	(9.0-14.2)	11.0	(9.0-12.7)	10.9	(9.0-12.7)	12.2	(10.0-14.5)	0.629**
TSF Adequacy 5° (%) ³	203.5	(153.0-240.0)	200.0	(184.6-244.4)	180.0	(140.0-240.0)	219.1	(181.1-234.1)	0.597**
MAMC (cm) ¹	13.8	±2.0	13.1	±0.5	14.2	±0.6	13.9	±0.5	0.433†
Adequacy 5° (%) ¹	95.9	±11.0	97.2	±3.6	95.9	±2.8	92.1	±3.4	0.310**

Note: Values expressed as: ¹Mean and standard deviation; ²Prevalence and confidence interval of 95%; ³Median and Interquartile Interval (25-75%). **Fisher's exact test; †Analysis of Variance (Anova) heterogeneity test; **Anova trend test; **Kruskal-Wallis test. BMI: Body Mass Index; TSF: Triceps Skinfold; MAMC: Mid-Arm Muscle Circumference.

(146.2±27.9%; p=0.033). However, when the TCV was assessed in categories, it was found that both groups of patients, heterozygous for delta F508 (5.9%; 95%CI=0.0-45.9) and homozygous for delta F508 (10.0%; 95%CI=0.4-64.1), had the lowest values of prevalence ≥150% EER when

compared to the non-delta F508 patients (44.4%; CI95%=11.8-88.2; p=0.026).

When the prevalence of consumption ≥150% EER was assessed between the groups with presence (homozygous and heterozygous) and absence of delta F508 mutation, lower

Table 3. Consumption of calories, macronutrients, and dietary fibers according to the presence of delta F508 mutation. *Florianópolis* (SC), Brazil, 2012-2013.

Variables	Total n=36	Non-delta F508 carriers n=9	Heterozygous for delta F508 n=17	Homozygous for delta F508 n=10	p-value
TCV (calories/day) ¹	1787.2 ±297.7	1884.9 ±243.8	1672.8 ±324.0	1893.9 ±241.3	0.089 [†]
TCV (% EER) ¹	127.1 ±30.2	146.2 ±27.9	115.0 ±32.5	130.6 ±17.8	0.033 [†]
<i>Total calorie value</i>					
<110% of EER (%) ²	36.1 (13.8-68.4)	11.1 (0.2-36.0)	58.8 (31.6-86.1)	20.0 (5.0-53.8)	
≥110% and <150% of EER (%) ²	47.2 (23.0-72.2)	44.4 (22.2-72.2)	35.3 (14.2-61.6)	70.0 (44.0-89.7)	0.026 ^{**}
≥150% of EER (%) ²	16.6 (0.4-64.1)	44.4 (11.8-88.2)	5.9 (0.0-45.9)	10.0 (0.4-64.1)	
<i>Carbohydrates</i>					
Grams/day ¹	218.2 ±35.2	236.4 ±31.3	204.1 ±34.2	225.7 ±32.8	0.056 [†]
% TCV (%) ¹	49.3 ±4.8	50.3 ±5.2	49.3 ±5.2	48.2 ±3.7	0.346 ^{**}
40-50% of TCV (%) ²	69.5 (46.5-85.0)	44.4 (24.4-65.1)	76.5 (54.8-90.6)	80.0 (59.3-93.2)	0.171 ^{**}
51-60% of TCV (%) ²	30.5 (6.0-61.0)	55.6 (23.4-83.2)	23.5 (6.0-61.0)	20.0 (2.3-51.7)	
<i>Proteins</i>					
Grams/day ¹	65.9 ±18.2	66.1 ±12.4	62.8 ±23.2	70.9 ±12.0	0.545 [†]
% TCV (%) ¹	14.7 ±2.8	14.0 ±2.2	14.8 ±3.4	15.2 ±2.4	0.351 ^{**}
10-15% of TCV (%) ²	72.2 (52.2-88.4)	77.8 (56.3-91.0)	70.6 (48.2-85.7)	70.0 (48.2-85.7)	1.00 ^{**}
16-25% do TCV (%) ²	27.8 (6.7-65.2)	22.2 (2.5-55.6)	29.4 (6.6-65.2)	30.0 (6.6-65.2)	
<i>Lipids</i>					
Grams/day ¹	72.8 ±16.0	74.8 ±16.6	68.2 ±16.5	78.8 ±13.5	0.233 [†]
% TCV (%) ¹	36.7 ±4.8	35.6 ±4.9	36.5 ±4.7	38.1 ±5.3	0.263 ^{**}
25-35% of TCV (%) ²	47.2 (23.0-72.2)	44.4 (23.0-72.2)	52.9 (27.8-77.0)	40.0 (18.4-67.1)	0.910 ^{**}
36-50% of TCV (%) ²	52.8 (28.9-75.5)	55.6 (33.5-79.7)	47.1 (22.4-71.1)	60.0 (33.5-79.7)	
Dietary fibers (grams/day) ¹	9.7 ±1.4	9.4 ±0.7	9.6 ±0.9	10.2 ±2.4	0.266 ^{**}

Note: Values expressed as: ¹Mean and standard deviation; ²Prevalence and confidence interval of 95%. ^{**}Fisher's exact test; [†]Analysis of Variance (Anova) heterogeneity test; ^{**}Anova trend test.

TCV: Total Caloric Value; EER: Estimated Energy Requirement.

Table 4. Poisson regression model: negative association for BMI ≥25°, TCV ≥150% EER, and presence of delta F508 mutation. *Florianópolis* (SC), Brazil, 2012-2013.

	¹ BMI ≥25°		² TCV ≥150% EER	
	PR (95%CI)	p-value*	PR (95%CI)	p-value*
Presence of delta F508 mutation	1		1	
Absence of delta F508 mutation	0.4 (0.2-0.8)	0.006	0.1 (0.02-0.3)	0.001

Note: ¹Adjusted for FEV, (%) and %EER (mean); ²Adjusted for BMI; *Wald test.

BMI: Body Mass Index; TCV: Total Caloric Value; EER: Estimated Energy Requirement; PR: Prevalence Ratio; 95%CI: 95% Confidence Interval.

prevalence of consumption was observed in the group with the mutation (7.4% versus 44.4%, Chi-square test, $p=0.010$) (data not included in the table). With regard to the macronutrient intake, only the carbohydrates showed a significant statistical difference between the groups, with higher consumption in the group of

patients with absence of delta F508 mutation (236.4 g/day ±31.3; $p=0.056$).

A Poisson regression model was developed to estimate the prevalence ratios for BMI ≥25° and TCV ≥150% EER between the groups with presence (homozygous and heterozygous for delta F508) and absence of delta F508 mutation

(Table 4). Among the children and adolescents with delta F508 mutation, there was 60% lower prevalence ($PR=0.4$; $95\%CI=0.2-0.8$) of $BMI \geq 25^\circ$ (adjusted for % FEV and % EER) and 90% lower prevalence ($PR=0.1$; $95\%CI=0.02-0.3$) of $TCV \geq 150\%$ EER (adjusted for BMI) when compared to the individuals not carrying this mutation.

DISCUSSION

In this study, a significant association between the low BMI percentile and the presence of delta F508 mutation was found. Patients homozygous for delta F508 had lower BMI percentile ($21.9 \pm 23.3^\circ$; $p=0.017$), value lower than that considered acceptable ($BMI \geq 25^\circ$), according to the American Consensus on Nutrition for cystic fibrosis¹⁸. This result corroborates other studies, which despite using other indicators (weight, height, weight and height percentile for age, and weight and BMI Z-scores) found values lower than those obtained in the present study when delta F508 mutation was present⁵⁻⁸. The latest recommendation for patients with cystic fibrosis is that the BMI should be maintained as far as possible above 50° ¹³, and in general, the sample average value was not close to that value ($33.0^\circ \pm 27.9$).

A significant association between the daily energy consumption above EER and the presence of delta F508 mutation was also observed in this study. Even though, on average, all patients' EER exceeded 110% ¹³, either the homozygous or heterozygous patients for delta F50 showed lower average percentages and lower prevalence $\geq 150\%$ EER when compared to non-delta F508 patients. It is known that as far as cystic fibrosis is concerned, there are many factors that hinder food intake (inflammation and chronic infections, use of medications, intestinal losses, and cough among others) and that energy requirement increase is individual^{13,18,26}. However, it is important to examine the type of genotype to

increase the energy requirement once delta F508 mutation has already been associated with increased energy expenditure *per se*. In studies carried out in Philadelphia⁹ and Australia¹⁰, the energy expenditure of cystic fibrosis children homozygous for delta F508 was 23 and 30% higher, respectively, when compared to healthy children of the control group.

In the final model of Poisson regression, it could be seen that the presence of delta F508 mutation was negatively and significantly associated with BMI and energy consumption. In general, patients carrying delta F508 showed lower caloric intake and, consequently, poorer nutritional status. Individuals with cystic fibrosis with good nutritional status also showed better health conditions, especially better lung function assessed by FEV_1 ⁸. With respect to the presence of delta F508 mutation, it has already been observed a protective effect of high BMI on FEV_1 in adolescents and children homozygous for said mutation²⁷.

Some results do not show significant association between the nutritional status^{28,29} and food intake^{9,28,30} in the presence of delta F508 mutation, which indicates that irrespective of the large existing variability, which is due to the genotype, environmental factors may also have influence (including beneficial and harmful effects of the treatment).

The type of design and the assessment of food intake can be considered limitations of the present study. Cross-sectional studies do not allow for the establishment of a causal relationship; therefore, the association between this study outcomes and the presence of delta F508 mutation may be subject to reverse causality. The limitation of the 3-day diet record is the fact that it can lead the individual to change choices and overestimate or underestimate the amount of foods. But such limitation was attenuated by the two methods of adjustment used in the study. The first was the within-person and between-person variance adjustment for the 3-day diet

records to remove the effect of variation of the individuals' food intake on themselves and on others²³. The second was the residual method of nutrients with the purpose of removing the variation caused by energy consumption²⁴.

Although this study had a small sample size, the number of individuals participating in this research corresponded to more than half of the eligible patients (of a total of 54 eligible individuals, data from 36 patients were collected) at the cystic fibrosis care center of the children hospital in the state of *Santa Catarina*, Brazil.

Therefore, it can be concluded that the analysis of possible associations with delta F508 mutation is not the answer for all phenotypes, but in the clinical context, a good characterization of cystic fibrosis children and adolescents is possible providing a better guidance and awareness of nutritional care.

Thus, close monitoring of the nutritional status and a specific dietary intervention should be part of the diet therapeutic approach to this disease aiming at improving the patients' overall health. Further cohort studies are needed for a better understanding of the associations found.

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CONTRIBUTORS

C SANTOS and T STEEMBURGO contributed to the conception and design of the study, data collection, interpretation, and analysis. All authors critically reviewed the manuscript and approved the final version submitted for publication.

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