Comparison of total body chlorine, potassium, and water measurements in children with cystic fibrosis[1–3]

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ABSTRACT

Background: Symptoms of cystic fibrosis (CF) may limit the utility of total body chlorine (TBCl) and total body potassium (TBK) measurements for assessing body fluid compartments of children.

Objective: This study assessed relations among independent measurements of TBCl, TBK, and total body water (TBW) in children with CF.

Design: We compared cross-sectional measurements of TBCl by in vivo neutron activation analysis, TBK by whole-body counting of ^{40}K, TBW by D_2O dilution [TBW(D_2O)], and TBW from TBCl and TBK [TBW(Cl + K)] in 19 prepubertal children (13 boys) aged 7.6–12.5 y who had mild symptoms of CF. Body-composition measurements were compared with data from previous studies of healthy children.

Results: Subjects with CF had deficits in TBCl, TBK, TBW, and body weight compared with control reference data ($P < 0.05$). The ratios (TBCl + TBK)/TBW and TBCl/TBK were not significantly different from control reference values, and plasma chlorine and potassium concentrations were within control reference ranges. The sum of TBCl and TBK correlated with TBW(D_2O) ($r^2 = 0.79$, $P < 0.001$), and TBW(Cl + K) correlated with TBW(D_2O) ($r^2 = 0.78$, $P < 0.001$). TBW(Cl + K) was similar to TBW(D_2O) ($\bar{x} \pm$ SEM: 19.0 ± 0.5 compared with 19.4 ± 0.5 L; NS).

Conclusions: Prepubertal children with mild symptoms of CF can develop deficits in TBCl, TBK, and TBW that reflect chronic energy malnutrition. Mild symptoms of CF do not appear to affect normal relations among TBCl, TBK, and TBW. Measurements of TBCl and TBK may be used to assess body fluid compartments in these patients. *Am J Clin Nutr* 2000;71:36–43.

KEY WORDS Body composition, total body chlorine, total body potassium, total body water, extracellular water, intracellular water, cystic fibrosis, prepubertal children

INTRODUCTION

Protein-energy malnutrition is a common sequela in children with cystic fibrosis (CF) (1) and may result in abnormal whole-body distributions of electrolytes and fluids (2–4). The assessment of whole-body electrolyte and fluid compartments is therefore important in determining the nutritional status of children with CF.

Measurements of total body chlorine (TBCl) and total body potassium (TBK) have been used to derive estimates of extra- and intracellular fluid compartments in normal-weight and obese adults (5, 6). This approach is based on the assumptions that ≈88% of TBCl is found in extracellular water (ECW) (7) and that >96% of TBK is found in intracellular water (ICW) (8).

In children with CF, TBCl and TBK measurements may be influenced by factors that include protein-energy malnutrition and the transepithelial chlorine transport abnormality that is the intrinsic defect in CF (9). Although TBCl, TBK, and fluid compartments were studied previously in children with CF (2–4, 10–12, and IRJ Humphries and KJ Gaskin, unpublished observations, 1996), interpretation of these data is problematic because of the small numbers of subjects, wide variations in disease severity, incomplete reporting of disease symptoms and body components, and use of body-composition methods that may be invalid in CF populations.

The aim of this study was to define body composition in a group of clinically stable prepubertal children with mild symptoms of CF by *1*) comparing independent measurements of TBCl, TBK, and total body water (TBW) and *2*) investigating the respective relations of TBCl and TBK to ECW and ICW.

SUBJECTS AND METHODS

Subject selection

The study group consisted of 19 children (13 boys) with CF who were aged 7.6–12.5 y. All subjects were selected from the CF clinic of the Royal Children's Hospital, Melbourne, according to the following criteria: *1*) age 7.0–13.0 y; *2*) prepubertal (ie, Tanner stages 1 or 2); *3*) positive diagnosis of CF on the basis of genotyping or pulmonary and gastrointestinal symptoms complemented by increased sweat chloride concentrations; *4*) mild

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symptoms of lung disease as judged by a physical examination, chest X-rays, and spirometry tests; *5*) no pulmonary exacerbations that required hospitalization within 2 mo of the study; *6*) no hospitalization within 2 mo of the study; *7*) weight change < 1.5 kg over the 2 mo before the study; *8*) no comorbidities likely to confound body-composition measurements (eg, clinical or biochemical evidence of liver disease or hepatosplenomegaly, pancreatitis, diabetes, or edema); and *9*) no history of long-term (> 1 y) use of medications likely to confound body-composition measurements (eg, corticosteroids, growth hormone, or diuretics). Reference data from previous studies of healthy children were used to interpret clinical, biochemical, and body-composition measures.

Study protocol

All subjects were studied as outpatients in 2 phases with the approval of the human ethics committees of both the Royal Children's Hospital and the Monash Medical Centre and the signed, witnessed, and informed consent of subjects and their parents or guardians. In phase 1, clinical status, diet, and pancreatic function were assessed. In phase 2, anthropometry, body composition, and plasma biochemistry were assessed within a 4-h morning session. Each subject participated in both phases within a period of ≤12 wk.

Clinical assessment

Body weight was measured with a digital scale and body height was determined by using a stadiometer according to the standards of Lohman et al (13). Anthropometric measures were interpreted by using World Health Organization (WHO) normal population percentiles for weight and height (14). Growth velocities were expressed as a percentage of the normal 50th-percentile growth velocities reported by Tanner et al (15), and pubertal status was determined according to the Tanner stages of development (16).

Medical records of spirometry test results were reviewed to determine each subject's measure of forced expiratory volume in 1 s (FEV_1), expressed as a percentage of that expected of healthy children of the same age and sex. The pulmonary status of each subject was assessed by a thoracic physician and assigned a Holzer score (17) as follows: 0 (no lung disease) for subjects with no persistent cough, no sputum production, a normal chest radiograph, and an FEV_1 > 80%; 1 (minimal lung disease) for subjects with persistent cough, no sputum production, a chest radiograph showing minimal streaking, and an FEV_1 > 60% but < 80%; and 2 (mild lung disease) for subjects with persistent cough, sputum production < 10 mL/d, a chest radiograph showing bronchial-wall thickening and minimal hyperinflation, and an FEV_1 > 65% but < 85%.

Subjects receiving pancreatic enzymes at the time of the study were considered to be pancreatic insufficient. Pancreatic status was also assessed by a 3-d fat-clearance study (18). Each subject completed a 3-d weighed-food diary and stool collection within 2 wk of body-composition assessment. The fat content of stools was assessed by using the method of Van der Kamer et al (19), and a coefficient of fat absorption was calculated as 100 × [fat intake (g) − fat excreted in stool (g)]/[fat intake (g)].

The nutrient contents of dietary food items were analyzed by using NUTRITIONIST III, version 4 (N-Squared Computing, Salem, OR; modified for use by Monash University, Melbourne, Australia) and food-composition tables (20). Energy and protein intakes were expressed as a percentage of the recommended dietary intakes (RDIs) published by the National Health and Medical Research Council of Australia (20). Medical histories were reviewed to determine each subject's use of corticosteroids.

Laboratory assessment

Subjects were designated as either homozygous or heterozygous for the ΔF508 mutation on the basis of DNA testing by the Victorian Clinical Genetics Service, Melbourne. Fasting venous blood samples were collected for laboratory analysis immediately before each subject's body-composition assessment.

Body-composition assessment

TBCl was measured by the method of prompt gamma-ray in vivo neutron activation analysis (IVNAA) as described elsewhere (21, 22). Subjects were irradiated unilaterally by a 0.2-GBq ^{252}Cf neutron source from shoulder to midthigh in both supine and prone positions. Prompt gamma-ray spectra were measured with 2 pairs of NaI(Tl) crystals (each crystal: 10 × 10 × 15 cm) positioned on both sides of the subject. TBCl was estimated from the ratio of chlorine-to-hydrogen counts as determined from the measurement of 8.57 and 2.22 MeV prompt gamma-rays from the respective reactions $^{35}Cl(n,\gamma)^{36}Cl$ and $^{1}H(n,\gamma)D$. The chlorine-to-hydrogen count ratio was corrected for the effect of body width and thickness on background and gamma-ray attenuation. Total body hydrogen was used as an internal standard, which was determined by using a 4-compartment model of body weight (22). The total exposure time per measurement scan was 630 s and the effective dose equivalent for a small child was 0.25 mSv (Q = 20, indicating exposure to neutrons). During this study, the assay error and precision were determined to be ≈3% and ≈10% (CV), respectively, for TBCl measurements.

TBK was measured by counting 1.46 MeV gamma-ray emissions from ^{40}K by using a shadow shield counter (23). Subjects rested supine on a moveable bed that passed through a symmetric array of four 40 × 10 × 10-cm NaI(Tl) detectors shielded within an open-ended, box-like steel chamber; the total scan time was 30 min. Measurements were calibrated against a potassium-rich, child-size box phantom, and a correction factor was applied to account for the effect of body thickness and width on gamma-ray attenuation (21). During this study, the error and precision of TBK measurements were determined to be ≈2.5% and ≈4% (CV), respectively.

TBW was measured by using the D_2O isotope dilution method (24). A baseline venous blood sample was first collected from the fasting subject and then an oral dose of ≈0.4 g 99.9% pure D_2O/kg body wt was administered with a mouthwash of deionized water (≈100 mL). The subject fasted for a further 2–3-h equilibration period until a second venous blood sample was collected. The D_2O plasma concentration of each plasma sample was determined in triplicate by using the Fourier transform infrared analysis technique and the average measure was then used to determine TBW by using the equation

$$TBW(D_2O)(L) = D_2O\ dose\ (g)/\ [(C_2 - C_1) \times 0.94 \times 0.95] \quad (1)$$

where C_1 is the plasma concentration of D_2O at baseline, C_2 is the D_2O plasma concentration after the equilibration period, 0.94 is a factor that corrects for the protein content of plasma, and 0.95 is a factor that corrects for the fraction of ^{2}H that exchanges with nonaqueous ^{1}H. The assay measurement error and precision for determining D_2O concentrations in spiked standard solutions were reported to be ≈1% and ≈2% (CV), respectively (25).

Reference values for the ratios (TBCl + TBK)/TBW and TBCl/TBK were determined from the data of Cheek (26). Selected reference constants were representative of healthy children aged 10 y. Measurements of TBCl, TBK, and TBW were compared with predictor equations derived from data reported by previous studies of healthy children (26–32). The predictor equations used were as follows:

$$\text{TBCl (g)} = 0.380 \times \text{age}^2 - 3.416 \times \text{age} + 38.23 \quad (2)\ (26\text{–}29)$$

$$\text{TBCl (g)} = 0.00806 \times \text{height}^2 - 1.528 \times \text{height} + 99.44 \quad (3)\ (26\text{–}29)$$

$$\text{TBK (g)} = 0.721 \times \text{age}^2 - 6.973 \times \text{age} + 66.940 \quad (4)\ (29)$$

$$\text{TBK (g)} = 0.0156 \times \text{height}^2 - 3.071 \times \text{height} + 195.8 \quad (5)\ (29)$$

$$\text{TBW (L)} = 1.986 \times \text{age} + 0.046 \quad (6)\ (30\text{–}32)$$

$$\text{TBW (L)} = 0.335 \times \text{height} - 26.089 \quad (7)\ (30\text{–}32)$$

where age is in years, height is in centimeters, and sex was found to be a nonsignificant predictor of TBCl, TBK, and TBW. The appropriateness of these equations is in part supported by the following criteria used to select the population reference data: *1*) only whites were included; *2*) only prepubertal children were included unless puberty per se did not significantly affect body composition; *3*) the weight and height of selected populations were representative of the WHO 50th-percentile values for weight-for-age, weight-for-height, and height-for-age (14); *4*) selected populations were assessed by body-composition methods similar to those used in this study; and *5*) selected population reference data allowed for comparisons on a sex-, age-, and height-standardized basis. Measurements of TBCl, TBK, and TBW were expressed as a percentage of the expected reference value for age or for height as determined by the aforementioned predictor equations.

ECW was derived from the measurement of TBCl as

$$\text{ECW (L)} = [\text{TBCl (mmol)} \times 0.88 \times 0.95 \times 0.94]/[\text{Cl}]_p\text{(mmol/L)} \quad (8)$$

where 0.88 is the assumed proportion of TBCl found in ECW, 0.95 accounts for the Donnan equilibrium factor, 0.94 accounts for the protein content of plasma, $[\text{Cl}]_p$ is the plasma chloride concentration measured in each subject, and $[\text{Cl}]_p$ is assumed to be representative of the average chlorine concentration of ECW (3, 7, 8).

ICW was derived from the measurement of TBK as

$$\text{ICW (L)} = \text{TBK (mmol)} \times (0.00833\ \text{kg/mmol}) \times 0.75\ \text{(L/kg)} \quad (9)$$

where 0.00833 kg/mmol is the ratio of body cell mass to TBK and 0.75 is the ICW content of the body cell mass (33, 34). This equation assumes that 100% of TBK is found within the body cell mass.

TBW was also derived from measurements of TBCl and TBK as

$$\text{TBW(Cl + K) (L)} = \text{ECW (L)} + \text{ICW (L)} \quad (10)$$

Statistical analysis

The one-sample *t* test was used to compare standardized measures of body composition, growth, pulmonary and gastrointestinal status, and dietary intake with the hypothesized control value of 100%. Measurements of weight, height, and body mass index (BMI) were compared with population reference data published by the WHO on the basis of *z* scores (14). For individual subjects, the normal reference range of *z* scores was interpreted as −2 to +2. The one-sample *t* test was used to compare the subjects' *z* scores with the hypothesized control *z* score of 0.

The Bland and Altman method of assessing agreement between 2 clinical methods (35) was used to compare TBW measurements determined by D_2O dilution with TBW measurements derived from the sum of TBCl and TBK. The paired *t* test was also used to compare absolute estimates of TBW(D_2O) with TBW(Cl + K).

Univariate linear regression analysis was used to assess relations between *1*) TBW(D_2O) and both TBW(Cl + K) and TBCl + TBK, *2*) continuous clinical variables and the measurement bias between TBW(D_2O) and TBW(Cl + K), and *3*) continuous clinical and laboratory variables on the one hand and body-composition variables on the other (GRAPHPAD PRISM, version 1.0; GraphPad Software, Inc, San Diego).

The unpaired *t* test was used to compare 2 subgroups of subjects (eg, boys compared with girls), and one-way analysis of variance with a Bonferroni correction was used to compare more than 2 subgroups of subjects (eg, genotype: *ΔF508/ΔF508* compared with ΔF508/other compared with other/other) (GRAPHPAD PRISM, version 1.0). An effect was assumed to be significant if $P < 0.05$.

RESULTS

Clinical assessment

Given that there were no significant sex differences in any of the clinical measures investigated, the clinical characteristics of boys and girls are combined in **Table 1**. The CF group had subnormal *z* scores for weight-for-age, weight-for-height, and BMI and a mean height *z* score indicative of a nonsignificant degree of stunting. Negative linear correlations were observed between age and *z* scores for weight-for-age ($r^2 = 32.2$, $P = 0.01$) and height-for-age ($r^2 = 37.0$, $P < 0.01$). Individual *z* scores for weight-for-age, weight-for-height, and height-for-age all fell within 2 SDs of reference medians. BMI-for-age *z* scores of all but 2 subjects fell within 2 SDs of the reference median.

Subjects showed a subnormal rate of growth in weight and a normal rate of growth in height. All subjects were prepubertal with Tanner scores of 1 or 2. Subjects had minimal symptoms of lung disease, as judged by a mean (±SEM) Holzer score of 0.9 ± 0.2, and subnormal lung function ($FEV_1 < 100\%$; $P < 0.001$). At the individual level, 6 subjects (all boys) showed no clinically overt symptoms of lung disease, 9 (5 boys) showed minimal symptoms of lung disease, and 4 (2 boys) showed mild symptoms of lung disease.

All subjects were pancreatic insufficient and used pancreatic-enzyme supplements. Dietary fat absorption tests were successfully completed for 15 of 19 subjects. These 15 subjects could absorb only 86.4% of ingested dietary fat with the use of enzyme supplements.

At the time of study, all subjects were consuming an ad libitum diet. The 3-d weighed-food diary was completed by 18 of 19 subjects. A comparison with RDIs showed a significant deficit in total energy intake and a significant surfeit of protein intake. Only 2 subjects (1 boy) reported taking salt-tablet supplements in quantities of ≈1000 mg/d over the 3-d period of dietary assessment.

TABLE 1
Clinical and laboratory assessment of children with cystic fibrosis[1]

Variable	Clinical or laboratory assessment
Age (y) (n = 19, 13 boys)	10.4 ± 0.4 (7.6–12.5)
Anthropometry (n = 19, 13 boys)	
Weight z score	−0.52 ± 0.20[2] (−1.48–1.88)
Height z score	−0.20 ± 0.21 (−1.41–1.87)
Weight-for-height z score	−0.61 ± 0.17[3] (−1.74– 0.72)
BMI z score	−0.67 ± 0.25[2] (−2.02–2.64)
Growth (n = 19, 13 boys)	
Weight velocity (% of ideal)	70 ± 7[4] (21–159)
Height velocity (% of ideal)	100 ± 4 (66–143)
Pubertal status (Tanner stage)	1.1 ± 0.1 (1–2)
Pulmonary status (n = 19, 13 boys)	
Holzer score	0.9 ± 0.2 (0–2)
FEV_1 (%)	83 ± 5[4] (44–127)
Gastrointestinal status	
Pancreatic insufficient (n = 19, 13 boys)	
Fat absorption (%) (n = 15, 10 boys)	86.4 ± 1.6[4] (74–94)
Diet (n = 18, 12 boys)	
Energy (% of RDI)	89 ± 4[5] (57–133)
Protein (% of RDI)	230 ± 14[4] (127–339)
Sodium ($mg \cdot kg^{-1} \cdot d^{-1}$)	89 ± 7 (50–145)
Potassium ($mg \cdot kg^{-1} \cdot d^{-1}$)	89 ± 6 (42–128)
Plasma electrolytes (n = 19, 13 boys)	
Chloride (mmol/L)	103.9 ± 0.3 (102–108)[6]
Sodium (mmol/L)	140.2 ± 0.6 (137–144)[7]
Potassium (mmol/L)	4.6 ± 0.1 (3.4–5.2)[8]

[1] $\bar{x}$ ± SEM; range in parentheses. FEV_1, forced expiratory volume in 1 s; RDI, recommended dietary intake (20).
[2,3] Significantly different from zero (one-sample t test): [2] $P < 0.05$, [3] $P < 0.01$.
[4,5] Significantly different from 100% (one-sample t test): [4] $P < 0.001$, [5] $P < 0.05$.
[6–8] Reference ranges: [6] 98–110 mmol/L, [7] 135–145 mmol/L, [8] 3.5–5.0 mmol/L.

Only 7 subjects (3 boys) had used steroids, ranging in frequency from once or twice in their lifetime through to several times/y. There were 17 d on which body-composition measurements were performed. The mean (±SD) maximum temperature for these 17 d was 22.3 ± 6.2 °C (range: 15.0–36.5).

Laboratory assessment

Seven subjects (5 boys) were homozygous for the *ΔF508* allele, 9 (6 boys) were heterozygous for the *ΔF508* allele, and 3 (2 boys) had no *ΔF508* allele. The CF group had plasma chlorine, sodium, and potassium concentrations within the control reference ranges. Plasma chlorine and potassium concentrations showed no significant association with age, sex, genotype expressed in terms of the *ΔF508* allele, steroid use, or any of the variables of anthropometry, growth, pulmonary status, gastrointestinal status, or diet.

Body-composition assessment

General effects of cystic fibrosis on body composition

The body composition of the 19 subjects studied is summarized in **Table 2**. Deficits were observed for both age- and height-standardized values of TBCl, TBK, and TBW. Negative linear correlations were observed between age and age-standardized values of TBCl (r^2 = 29.2, P = 0.02), TBK (r^2 = 30.6, P = 0.01), and TBW (r^2 = 41.2, $P < 0.01$) considered separately. By contrast, mean values for the ratios (TBCl + TBK)/TBW and TBCl/TBK did not differ significantly from the cited reference values. Neither of these ratios was significantly associated with age, genotype expressed in terms of the *ΔF508* allele, steroid use, or any of the variables of anthropometry, growth, pulmonary status, gastrointestinal status, or diet. Sex did not significantly affect any body-composition measure.

Comparison of TBCl and TBK with TBW(D_2O)

There was a relatively strong linear correlation between the sum of TBCl and TBK and TBW(D_2O), as shown in **Figure 1**. The sum of TBCl and TBK was a significant predictor of TBW(D_2O) ($P < 0.001$), accounting for 79% of the variation in TBW(D_2O).

Comparison of TBW(Cl + K) with TBW(D_2O)

Absolute mean values of TBW(D_2O) and TBW(Cl + K) in boys and girls and in the combined group are shown in **Table 3**. The paired t test showed no significant difference between the 2 estimates of TBW. In addition, no significant sex difference was found in estimates of TBW by either method when the unpaired t test was used.

A linear regression analysis of TBW(D_2O) compared with TBW(Cl + K) in the CF group showed TBW(Cl + K) to be a significant predictor of TBW(D_2O) ($P < 0.001$). The slope of the regression line was not significantly different from 1.0 (one-sample t test), and the intercept was not significantly different from 0 (one-sample t test).

Results from the Bland and Altman analysis of agreement between the D_2O dilution method and the method of summing TBCl and TBK are summarized in **Table 4** and **Figure 2**. The 95% CI for the average measurement bias between the 2 methods included zero for both boys and girls and for the combined group. For individual subjects, the variation of differences between the 2 methods are indicated by the limits of agreement, which spanned 4.95 L in boys, 2.87 L in girls, and 4.69 L in the combined group. Also, measurement biases between the 2 methods were not significantly associated with age, sex, genotype

TABLE 2
Body-composition assessment of children with cystic fibrosis[1]

Variable	Assessment
TBCl (mmol) (n = 19, 13 boys)	1097 ± 34 (824–1362)
Percentage TBCl for age (%)	88.2 ± 2.9[2] (63.5–113.4)
Percentage TBCl for height (%)	92.4 ± 2.5[3] (68.1–118.2)
TBK (mmol) (n = 19, 13 boys)	1717 ± 51 (1269–2059)
Percentage TBK for age (%)	91.7 ± 2.1[2] (81.1–114.3)
Percentage TBK for height (%)	94.9 ± 1.5[3] (84.6–105.6)
TBW[5] (L) (n = 19, 13 boys)	19.4 ± 0.5 (15.6–23.8)
Percentage TBW for age (%)	94.9 ± 2.4[4] (81.5–121.3)
Percentage TBW for height (%)	95.5 ± 1.4[3] (86.2–107.2)
(TBCl + TBK)/TBW[5] (mmol/L)	145.3 ± 1.9[6] (126.2–155.7)
TBCl/TBK (mmol/mmol)	0.64 ± 0.01[7] (0.49–0.79)

[1] $\bar{x}$ ± SEM; range in parentheses; CF, cystic fibrosis; TBCl, total body chlorine; TBK, total body potassium; TBW, total body water.
[2–4] Significantly different from 100% (one-sample t test): [2] $P < 0.001$, [3] $P < 0.01$, [4] $P < 0.05$.
[5] Measured by D_2O dilution.
[6,7] Not significantly different from reference values: [6] 146 mmol/L, [7] 0.66 mmol/mmol.

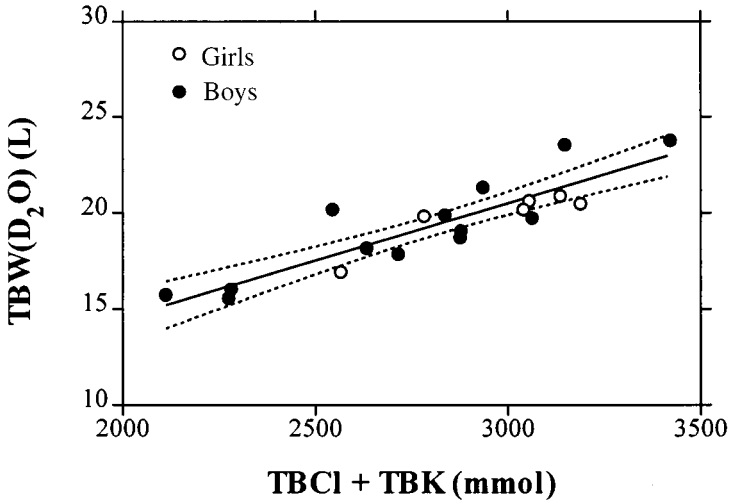

FIGURE 1. Total body water as measured by D_2O dilution [TBW(D_2O)] compared with the sum of total body chlorine plus total body potassium (TBCl + TBK) in 19 children (13 boys) with cystic fibrosis. Regression equation: TBW(D_2O) = 0.00594 *x* + 2.67 (SEE = 1.09 L, r^2 = 0.79, $P < 0.001$).

expressed in terms of the *ΔF508* allele, steroid use, or any of the variables of anthropometry, growth, pulmonary status, gastrointestinal status, or diet.

DISCUSSION

Effect of mild symptoms of cystic fibrosis on TBCl, TBK, and TBW

The CF group in this study had a body-composition pattern consistent with compromised growth of new tissue and loss of existing tissue. This was evidenced by *1*) significant deficits in age- and height-standardized measures of TBCl, TBK, TBW, and body weight; *2*) age-standardized deficits in TBCl, TBK, TBW, and body weight that increased significantly with age; and *3*) a subnormal weight velocity. By contrast, individual subjects appeared capable of attaining normal TBCl, TBK, TBW, or body weight for age or height.

There is a paucity of TBCl data from subjects with CF. Measurements of TBCl in apparently well-nourished children with CF were recorded (IRJ Humphries and KJ Gaskin, unpublished observations, 1996). TBCl was determined by IVNAA in 55 fibrocystic children with an average age of 10.8 y and 28 healthy children with an average age of 11.1 y. Overall, the CF group weighed 8 kg less and had 8% less TBCl than did the control group. However, when TBCl was adjusted for body weight, there was no significant difference in TBCl between the 2 groups. Humphries and Gaskin concluded that, on a body-weight basis, well-nourished children with CF have normal amounts of TBCl. The only other report of TBCl in subjects with CF appears to be that of Cheek (4), who used the NaBr dilution technique to measure TBCl in 5 malnourished, fibrocystic children aged between 0.5 and 4 y. Four of the 5 children had an increased ratio of TBCl to body weight; this finding was interpreted to signify a loss of total body fat. The TBCl observations in the present study extend the findings of Cheek (4) and are consistent with the data of Humphries and Gaskin. The TBK and TBW observations in this study support results of past studies showing that children with CF can attain normal amounts of TBK and TBW and that age- and height-related deficits in TBK and TBW develop in relation to the severity of CF symptoms (10–12, 36–38).

Multiple factors may affect TBCl, TBK, and TBW in children with CF. These include the normal influences of age, sex, growth, and physical activity (8, 26, 29–32, 39, 40) and the abnormal influences of malnutrition secondary to pulmonary and gastrointestinal symptoms of CF (12, 41, 42) and inadequate dietary intake (1, 43, 44). In this study, CF-related lung disease, pancreatic insufficiency, and an inadequate dietary energy intake were the factors most likely to influence body-composition abnormalities. This statement is supported by the observation that 18 of 19 subjects had minimal or mild symptoms of lung disease. Also, all subjects were pancreatic insufficient, and the CF group reported a reduced dietary energy intake equivalent to 89% of the RDI (*see* Table 1).

Comparisons between TBCl, TBK, and TBW

The results of this study suggest that children with mild symptoms of CF can maintain normal relations among TBCl, TBK, and TBW. This is supported by the following findings. First, a positive linear correlation was observed between TBCl + TBK and TBW(D_2O) (*see* Figure 1). This result was indepen-

TABLE 3
Total body water (TBW) as measured by D_2O dilution [TBW(D_2O)] and from the sum of total body chlorine plus total body potassium [TBW(Cl + K)] in children with cystic fibrosis[1]

	TBW(D_2O)	TBW(Cl + K)
	L	
Boys (*n* = 13)	19.2 ± 0.7 (15.6–23.8)	18.5 ± 0.7 (14.4–22.8)
Girls (*n* = 6)	19.8 ± 0.6 (16.9–20.9)	20.1 ± 0.7 (17.4–21.7)
Combined[2] (*n* = 19)	19.4 ± 0.5 (15.6–23.8)	19.0 ± 0.5 (14.4–22.8)

[1] $\bar{x}$ ± SEM; range in parentheses.
[2] Nonsignificant sex difference in TBW by either method, $P > 0.05$ (unpaired *t* test).

TABLE 4
Comparison of total body water (TBW) measured by D_2O dilution [TBW(D_2O)] with TBW estimated from the sum of total body chlorine plus total body potassium [TBW(Cl + K)] in children with cystic fibrosis[1]

TBW(Cl + K) − TBW(D_2O)	Boys (n = 13)	Girls (n = 6)	Combined (n = 19)
Bias (L)	−0.66	0.29	−0.36[1]
95% CI (L)[2]	−1.41, 0.09	−0.46, 1.04	−0.93, 0.20
Limits of agreement (L)[3]	−3.14, 1.81	−1.15, 1.72	−2.71, 1.98

[1] Nonsignificant sex difference (unpaired t test): $P > 0.05$.
[2] (Bias − 2 SEM) to (bias + 2 SEM).
[3] (Bias − 2 SD) to (bias + 2 SD).

dent of sex. Because TBCl is largely confined to the extracellular compartment (≈88% by weight) (3, 7), TBCl should be related to the volume of this compartment. Similarly, TBK is largely confined to the intracellular compartment (>96% by weight) (7, 33) and should therefore be related to the volume of this compartment. The observed correlation is not unexpected and is consistent with similar findings reported in healthy children of similar age (26) and in healthy adults (5).

Second, the ratio (TBCl + TBK)/TBW(D_2O) in the CF group attained a mean (±SEM) value of 145.3 ± 1.9 mmol/L, which was similar to the control value of 146 mmol/L calculated from the reference data of Cheek (26). Although this ratio showed wide variability in individual subjects (126.2–155.7 mmol/L), the ratio was not significantly associated with age, sex, or the measures of anthropometry, growth, genotype, pulmonary and gastrointestinal status, diet, or steroid use.

Third, the ratio TBCl/TBK was not significantly different from the control value derived from the reference data of Cheek (*see* Table 2). Our results are consistent with those of past studies of healthy humans that showed the ratio TBCl/TBK falling within a relatively constant range from ≈0.6 to ≈0.8 mmol/mmol during childhood, adolescence, and adulthood (26, 28).

The relation of TBCl to ECW

In healthy children and adults, ≈88% of TBCl is located in disparate pools of extracellular fluid (3, 7). Although chlorine concentrations of extracellular fluids may vary considerably, the average chlorine concentration of the total ECW pool is closely approximated by the chlorine concentration of blood plasma (≈100 mmol/L).

The intrinsic defect identifying CF is abnormal transepithelial chlorine transportation. Whether this defect is associated with TBCl abnormalities in CF patients was questioned previously. The results of the present study suggest that children with mild symptoms of CF can maintain a normal relation between TBCl and ECW. Subjects showed a normal plasma chlorine concentration consistent with that reported in previous studies (45–47). This study also found a strong linear correlation and a good agreement between TBW(Cl + K) and TBW(D_2O). In calculating ECW from TBCl, we assumed that 88% of TBCl is found in ECW and that the plasma chlorine concentration is representative of the average chlorine concentration of the total ECW pool. Our results suggest that these assumptions are suitable for clinically stable children with mild symptoms of CF.

The relation of TBK to ICW

This study identified the following supportive evidence for a normal relation between TBK and ICW in clinically stable children with mild symptoms of CF. First, plasma potassium concentrations were within the normal reference range for 17 of 19 subjects, slightly above the upper reference value for one subject (5.2 compared with 5.0 mmol/L), and slightly below the lower

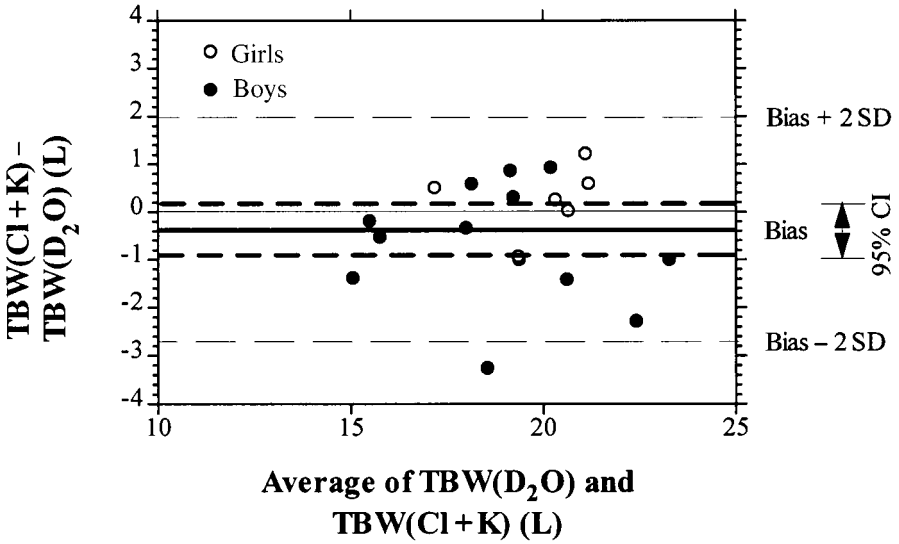

FIGURE 2. Differences between total body water measured by D_2O dilution [TBW(D_2O)] and TBW estimated from the sum of total body chlorine plus total body potassium [TBW(Cl + K)] in 19 children (13 boys) with cystic fibrosis. The mean difference (bias), the 95% CI of the mean difference, and the limits of agreement are also shown.

reference value for another (3.4 compared with 3.5 mmol/L).

Second, we observed a strong linear correlation and a good agreement between TBW(Cl + K) and TBW(D_2O). ICW was derived from TBK by using a cellular model that assumes that 100% of TBK is found in the body cell mass, that the ratio of body cell mass to TBK is 0.00833 kg/mmol, and that 75% of the body cell mass is ICW by weight (33, 34). Our data suggest that this cellular model of TBK is also suitable in children with mild symptoms of CF.

The validity of TBW measurements by D_2O dilution

This study used the D_2O dilution method as a reference standard against which TBW estimates derived from TBCl + TBK were compared. In an ancillary experiment performed in the CF group, no significant difference was observed between TBW measured by D_2O dilution and TBW derived from the 4-compartment model of body composition defined as body weight (measured by using digital scales) minus the sum of total body fat measured by dual-energy X-ray absorptiometry, total body protein determined by IVNAA, and total body bone mineral measured by dual-energy X-ray absorptiometry [$\bar{x} \pm$ SEM: 19.4 ± 0.5 compared with 19.6 ± 0.5 L for TBW(D_2O) and TBW(4-compartment model), respectively; $P = 0.36$ by paired t test] (21, 22). The significance of this comparison is that the 4-compartment model is based on methods that are independent of the primary CF defect.

Summary

This study showed independent, cross-sectional measurements of TBCl, TBK, and TBW in a group of clinically stable prepubertal children with mild symptoms of CF. Modest deficits in TBCl, TBK, and TBW were consistent with the effect of chronic energy malnutrition secondary to multiple factors that included CF lung disease, pancreatic insufficiency, and an energy-deficient diet. By contrast, relations among TBCl, TBK, and TBW appeared normal and unaffected by symptoms of CF. The clinical significance and implications of these findings are as follows. First, the abnormal CF gene that affects electrolyte and fluid transport across the epithelial surfaces of specific tissues and organs does not appear to affect to any significant extent normal relations between electrolytes and water compartments at a whole-body level. Second, noninvasive measurements of TBCl and TBK may be used as surrogate measures of ECW, ICW, and TBW in clinically stable children with mild symptoms of CF.

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REFERENCES

1. Roulet M. Protein-energy malnutrition in cystic fibrosis. Acta Paediatr Scand Suppl 1994;83:43–8.
2. Levy LD, Durie PR, Pencharz PB, Corey ML. Effects of long-term nutritional rehabilitation on body composition and clinical status in malnourished children and adolescents with cystic fibrosis. J Pediatr 1985;107:225–30.
3. Cheek DB. Extra-cellular volume: its structure and measurement and the influence of age and disease. J Pediatr 1961;58:103–25.
4. Cheek DB. Total body chloride of children in potassium deficiency and under circumstances of poor nutrition. Pediatrics 1954;14:193–9.
5. Yasumura S, Cohn SH, Ellis KJ. Measurement of extra-cellular space by total body neutron activation. Am J Physiol 1983;244:R36–40.
6. Mitra S, Plank LD, Knight GS, Hill GL. In vivo measurement of total body chlorine using the 8.57 MeV prompt de-excitation following thermal neutron capture. Phys Med Biol 1993;38:161–72.
7. Edelman IS, Leibman J. Anatomy of body water and electrolytes. Am J Med 1959;27:256–77.
8. Forbes GB. Human body composition: growth, aging, nutrition, and activity. New York: Springer-Verlag, 1987.
9. Bines JE. Body composition measurement: the challenge in the unwell child. Asian Pac J Clin Nutr 1995;4:39–42.
10. Shepherd RW, Holt TL, Greer R, Cleghorn GJ, Thomas BJ. Total body potassium in cystic fibrosis. J Pediatr Gastroenterol Nutr 1989;9:200–5.
11. Gaskin KJ, Waters DLM, Soutter VL, et al. Body composition in cystic fibrosis. In: Yasumura S, Harrison JE, McNeill KG, et al, eds. In vivo body composition studies: recent advances. New York: Plenum Press, 1990:15–21.
12. Thomas MA, Quirk P, Swanson CE, et al. Nutritional growth retardation is associated with defective lung growth in cystic fibrosis: a preventable determinant of progressive pulmonary dysfunction. Appl Nutr Invest 1995;11:350–4.
13. Lohman TG, Roche AF, Martorell R, eds. Anthropometric standardization reference manual. Champaign, IL: Human Kinetics, 1988.
14. World Health Organization. Measuring change in nutritional status. Geneva: WHO, 1983.
15. Tanner JM, Whitehouse RH, Takaishi M. Standards from birth to maturity for height, weight, height velocity, and weight velocity: British children, 1965. Part II. Arch Dis Child 1966;41:613–35.
16. Tanner JM. Growth at adolescence. 2nd ed. Oxford, United Kingdom: Blackwell Scientific Publications, 1962.
17. Holzer FJ, Olinsky A, Phelan PD. Variability of airways, hyper-activity and allergy in cystic fibrosis. Arch Dis Child 1981;56:455–9.
18. Anderson CM, Burke V, Gracey M, eds. Paediatric gastroenterology. 2nd ed. Melbourne: Blackwell, 1987:870–2.
19. Van der Kamer JH, ten Bokkel Huinink H, Weyers HA. Rapid method for the determination of fat in faeces. J Biol Chem 1949;177:347–55.
20. Rogers J. What food is that? and How healthy is it? Adelaide, Australia: Griffin Press, 1990.
21. Borovnicar D. The effect of cystic fibrosis on total body chlorine in children. PhD thesis. Monash University, Melbourne, 1998.
22. Borovnicar DJ, Stroud DB, Wahlqvist M, Strauss BJG. A neutron activation analysis facility for in vivo measurement of nitrogen and chlorine in children. Australas Phys Eng Sci Med 1996;19:252–63.
23. Bisnuth C, Elford M, Van Prooyen G. Calibration of a shadow shield detector. Instrumental Science Report. Melbourne, Australia: Department of Physics, Swinburne Institute of Technology, 1992.
24. Blagojevic N, Allen BJ, Gaskin KJ, Baur LA. Determination of total body water by Fourier transform infrared analysis. Australas Phys Eng Sci Med 1990;13:110–6.
25. Xiong DW. The effects of growth hormone on body composition in adults with growth hormone deficiency: with special reference to the study of total body water measurement. PhD thesis. Monash University, Melbourne, 1997.
26. Cheek DB, ed. Human growth. Philadelphia: Lea and Febiger, 1968.
27. Camerer W, Söldner. Die chemische Zusammensetzung des Neugeborenen. (The chemical composition of the newborn.) Z Biol 1900;39:173–92 (in German).
28. Ellis KJ. Reference man and woman more fully characterised: variations on the basis of body size, age, sex and race. Biol Trace Elem Res 1990;27:385–400.
29. Forbes GB. Growth of the lean body mass in man. Growth 1972;36:325–38.
30. Cheek DB, Mellits D, Elliot D. Body water, height and weight during growth in normal children. Am J Dis Child 1966;112:312–7.

31. Young CM, Bogan AD, Roe DA, Lutwak L. Body composition of preadolescent and adolescent girls. J Am Diet Assoc 1968;53:579–87.
32. Haschke F. Part II. Body composition of the male reference adolescent. Acta Paediatr Scand 1983;307:13–23.
33. Moore FD, Olesen KH, McMurrey D, et al., eds. The body cell mass and its supporting environment: body composition in health and disease. Philadelphia: WB Saunders, 1963.
34. Bruce Å, Andersson M, Arvidsson B, Isaksson B. Body composition. Prediction of normal body potassium, body water and body fat in adults on the basis of body height, body weight and age. Scand J Clin Lab Invest 1980;40:461–73.
35. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1986;1:307–10.
36. Tomezsko JL, Scanlin TF, Stallings VA. Body composition of children with cystic fibrosis with mild clinical manifestations compared with normal children. Am J Clin Nutr 1994;59:123–8.
37. Azcue M, Fried M, Pencharz PB. Use of bioelectrical impedance analysis to measure total body water in patients with cystic fibrosis. J Pediatr Gastroenterol Nutr 1993;16:440–5.
38. Newby MJ, Keim NL, Brown DL. Body composition of adult cystic fibrosis patients and control subjects as determined by densitometry, bioelectrical impedance, total-body electrical conductivity, skinfold measurements, and deuterium oxide dilution. Am J Clin Nutr 1990;52:209–13.
39. Fomom SJ, Haschke F, Ziegler EE, Nelson SE. Body composition of reference children from birth to age 10 years. Am J Clin Nutr 1982;35:1169–75.
40. Cheek DB. Observations on total body chlorine in children. Pediatrics 1954;14:5–10.
41. Kraemer R, Rüdeberg A, Hadorn B, Rossi E. Relative underweight in cystic fibrosis and its prognostic value. Acta Paediatr Scand 1978;67:33–7.
42. Lapey A, Kattwinkel J, Di Sant'Agnese PA, Laster L. Steatorrhea and azotorrhea and their relation to growth and nutrition in adolescents and young adults with cystic fibrosis. J Pediatr 1974;84:328–34.
43. Simopoulos AP, Bartter FC. The metabolic consequences of chloride deficiency. Nutr Rev 1980;38:201–5.
44. MacDonald A. Nutritional management of cystic fibrosis. Arch Dis Child 1996;74:81–7.
45. Lake CR, Davis PB, Ziegler M, Kopin IJ. Electrolyte and norepinephrine levels in blood of patients with cystic fibrosis. Clin Chim Acta 1979;92:141–6.
46. Aladjem M, Lotan D, Boichis H, et al. Renal function in patients with cystic fibrosis. Nephron 1983;34:84–6.
47. Miller ME, Cosgriff JE, Schwartz RH. Sweat bromide excretion in cystic fibrosis. J Lab Clin Med 1986;108:406–10.