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Body composition in patients with classical homocystinuria: body mass relates to homocysteine and choline metabolism

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ABSTRACT

Introduction: Classical homocystinuria is a rare genetic disease caused by cystathionine β-synthase deficiency, resulting in homocysteine accumulation. Growing evidence suggests that reduced fat mass in patients with classical homocystinuria may be associated with alterations in choline and homocysteine pathways. This study aimed to evaluate the body composition of patients with classical homocystinuria, identifying changes in body fat percentage and correlating findings with biochemical markers of homocysteine and choline pathways, lipoprotein levels and bone mineral density (BMD) T-scores.
Methods: Nine patients with classical homocystinuria were included in the study. Levels of homocysteine, methionine, cysteine, choline, betaine, dimethylglycine and ethanolamine were determined. Body composition was assessed by bioelectrical impedance analysis (BIA) in patients and in 18 controls. Data on the last BMD measurement and lipoprotein profile were obtained from medical records.
Results: Of 9 patients, 4 (44%) had a low body fat percentage, but no statistically significant differences were found between patients and controls. Homocysteine and methionine levels were negatively correlated with body mass index (BMI), while cysteine showed a positive correlation with BMI ($p < 0.05$). There was a trend between total choline levels and body fat percentage ($r = 0.439$, $p = 0.07$). HDL cholesterol correlated with choline and ethanolamine levels ($r = 0.757$, $p = 0.049$; $r = 0.847$, $p = 0.016$, respectively), and total cholesterol also correlated with choline levels ($r = 0.775$, $p = 0.041$). There was no association between BMD T-scores and body composition.
Conclusions: These results suggest that reduced fat mass is common in patients with classical homocystinuria, and that alterations in homocysteine and choline pathways affect body mass and lipid metabolism.

1. Introduction

Homocysteine is a toxic amino acid formed from methionine. High levels of homocysteine are associated with an increased incidence of several diseases, such as stroke, heart failure, coronary heart disease, dementia, and bone fractures (Homocysteine Studies Collaboration, 2002; Mudd et al., 1985). There are three main pathways by which homocysteine can be removed from plasma. In the transsulfuration pathway, homocysteine is irreversibly degraded by the action of the enzyme cystathionine beta-synthase (CβS; EC 4.2.1.22). It can also be remethylated by the ubiquitous methionine synthase (MS; EC 2.1.1.13), an enzyme dependent on vitamin B12 and folate, or by the liver/kidney specific betaine–homocysteine methyltransferase (BHMT; EC 2.1.1.5) using betaine. Betaine can be either derived from the diet or formed by oxidation of choline, a key nutrient in lipid metabolism.

Abbreviations: BIA, bioelectrical impedance analysis; BMD, bone mineral density; BMI, body mass index; CβS, cystathionine beta-synthase; DXA, dual-energy X-ray absorptiometry; ESPEN, European Society for Clinical Nutrition and Metabolism; HCPA, Hospital de Clínicas de Porto Alegre; HPLC, high performance liquid chromatography; IQ, interquartile range; SPSS, Statistical Package for the Social Sciences.

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Classical homocystinuria (OMIM 236200) is an autosomal recessive inborn error of metabolism caused by a deficiency in CβS, which results in increased plasma levels of homocysteine and methionine and decreased cysteine levels. It is a rare disease, with a worldwide prevalence estimated at 1:344,000 individuals (Mudd et al., 2001). Treatment includes pharmacological doses of pyridoxine (CβS cofactor), folic acid, vitamin B12, and, in some cases, betaine and also a methionine-restricted diet (Schiff and Blom, 2012). A large study on the natural history of the disease described equal proportions of patients responsive and unresponsive to pyridoxine (Mudd et al., 1985).

In addition to the classic manifestations (ectopia lentis, thromboembolism, mental retardation, and osteoporosis), patients with classical homocystinuria are tall and have a lean biotype (Brenton et al., 1972; Mudd et al., 1985). Until recently, it was believed that bone abnormalities were responsible for this phenotype. However, growing evidence suggests that these patients have reduced fat mass, associated with alterations in choline and homocysteine pathways.

In an animal model of classical homocystinuria, a marked decrease in adipose tissue was described as being associated with low levels of cysteine (Gupta and Kruger, 2011). Betaine and choline have also been associated with body composition, weight gain and lipid metabolism, both in healthy individuals and in experimental studies (Konstantinova et al., 2008; Teng et al., 2012; Wu et al., 2012). Moreover, there is evidence that choline and homocysteine metabolisms may overlap with respect to their relation to body weight (Zeisel, 2012). Given that the amount of body fat is closely related to bone mineral density (BMD), these changes could have important clinical implications in classical homocystinuria (Reid, 2008).

Despite the evidence from animal studies, this has not been studied in patients with classical homocystinuria. The objective of this study was to evaluate the body composition of patients with classical homocystinuria, identifying changes in body fat percentage and correlating findings with biochemical markers of homocysteine and choline pathways, lipoprotein levels and BMD T-scores.

2. Subjects and methods

The present study was approved by the Research Ethics Committee of Hospital de Clínicas de Porto Alegre (HCPA), Brazil, and the procedures were conducted after written informed consent was obtained from all subjects or their caretakers.

2.1. Study sample

Nine Brazilian patients with classical homocystinuria from 7 unrelated families were included in the study (median age = 26 years; IQ25–75 = 21–28 years). All patients had a late diagnosis (median age = 9 years; IQ25–75 = 6.25–12 years); 4 patients (44%) already had at least 3 systems compromised at diagnosis. Parental consanguinity was reported by 3/9 (33.3%) families.

At the time of study inclusion, all patients (aging 17–35 years) were receiving some type of treatment: pyridoxine (n = 7), folic acid (n = 8), betaine (n = 8), acetylsalicylic acid (n = 8), dietary methionine restriction (n = 9), and supplementation with a methionine-free formula (n = 2). However, most patients had not achieved adequate metabolic control (Table 1). Seven patients were unresponsive to pyridoxine, one was partially responsive (patient #4), and one was responsive (patient #3).

In addition, 18 healthy subjects (volunteers recruited from the institution) matched for gender and age, served as controls for bioelectrical impedance analysis (BIA) only. The controls had a median age of 25 years (IQ25–75 = 21–30 years).

The levels of homocysteine and methionine in the last 5 years (cysteine was unavailable) were obtained for 7 patients. For patient #7 these values were unavailable. Because patient #9 had a recent diagnosis, 3-year results of homocysteine and methionine measurements were obtained. Data on the last BMD measurement (T-score at the lumbar spine and femur), lipoprotein profile (triglycerides and HDL, LDL and total cholesterol) and clinical history were obtained from medical records. All patients had their diagnosis of classical homocystinuria based on the coexistence of hypermethioninemia and/or hyperhomocysteinemia and a positive cyanide-nitroprusside test, in addition to a clinical picture compatible with classical homocystinuria.

2.2. Assessment of body composition

Body composition was assessed in patients and controls in a single appointment by means of BIA (Biodynamics, 310e, Biodynamics Inc., Seattle, USA). Weight and height were measured and used to calculate BMI. BIA was performed using the tetrapolar method and following the recommendations of the European Society for Clinical Nutrition and Metabolism (ESPEN) (Kyle et al., 2004b). Based on the results obtained, body fat percentage was classified according to the cut-off points established by Heyward and Wagner (2004).

2.3. Assessment of BMD by dual-energy X-ray absorptiometry

BMD was assessed at the lumbar spine (L1–L4) and proximal and total femur by dual-energy X-ray absorptiometry (DXA) (GE—Lunar Prodigy, USA) at HCPA Department of Radiology. BMD was expressed as T-scores.

2.4. Laboratory assessment

Fasting blood was collected in EDTA tubes on the same day as BIA and anthropometry. Plasma was separated after centrifugation at 3000 rpm for 15 min. Total homocysteine, methionine and cysteine

Table 1
Results of the biochemical assessment and BMD of patients with classical homocystinuria (n = 9).

Patient	Current age (years)	Hcy (μmol/L)	Met	Cys	Free betaine (uM)	Free choline	Total choline	Free ethanolamine	Total ethanolamine	Free DMG	T score − BMD Spine	Femur
1a	31	321.73	593.30	124.97	12.2	4.81	297	7.28	14.4	3.89	−2.6	−1.9
1b	35	186.64	88.50	354.63	229.5	9.34	208	9.55	8.7	112	0.9	−0.9
1c	26	322.23	630.50	138.82	19.2	5.53	209	8.14	12.5	2.38	−1.4	−1.3
2	22	109.76	624.60	226.49	174	10.8	295	7.9	11.8	37.75	−0.5	−0.8
3	18	10.82	110.30	354.63	31.9	8.97	216	8.76	17.0	2.87	−1.3	NA
4	17	42.71	26.08	390.81	497.5	6.31	195	7.33	11.1	146.5	−1.4	0.2
5	21	233.86	915.03	206.93	432	12.4	184	8.81	9.9	81.5	−4.5	−2.4
6	28	48.65	69.20	349.62	585	9.75	322	7.05	16.4	53	NA	NA
7	26	66.10	29.0	370.43	49.9	5.60	218	6.29	11.7	5.2	NA	NA

Hcy: homocysteine, Met: methionine. Cys: cysteine, NA: data not available, BMD: bone mineral density, DMG: dimethylglycine.
Reference values of: Hcy: 5–15 μmol/L; Met: 5–30 μmol/L; Cys: 174–378 μmol/L (Skovby, 2003). Hcy target values for the treatment of classical homocystinuria are <20 μmol/L for pyridoxine-responsive patients and <60 μmol/L for the remaining patients (Wilcken, 2006).

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