原著論文

Resting energy expenditure and regional body composition in myotonic dystrophy type 1 patients

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ABSTRUCT

Objectives: In this study, we compared regional body composition and resting energy expenditure (REE) in myotonic dystrophy type 1 (DM1) patients with matched controls. We evaluated the relationship between regional body composition and REE in DM1 patients.

Methods: REE was studied by indirect calorimetry, and fat mass and fat-free mass were calculated by dual-energy X-ray absorptiometry in 18 DM1 patients and 10 age-matched healthy volunteers. In DM1 patients, we evaluated body muscle computed tomography and blood examination.

Results: DM1 patients had lower fat-free mass than controls ($P < 0.05$ - 0.001), but there were no group differences in age, height, weight, or fat mass. REE was lower in DM1 patients than controls ($P < 0.01$). In a multiple linear regression analysis, REE correlated with fat-free mass (beta 0.805, $P < 0.001$) and serum gamma glutamyl transpeptidase (beta 0.352, $P = 0.01$) in DM1 patients. In controls, REE correlated with weight (beta 0.86, $P = 0.001$).

Conclusion: REE decreased in DM1 patients. REE is proportional to whole body fat-free mass and serum gamma glutamyl transpeptidase in DM1 patients. Thus, REE should be a good indicator for evaluating new therapies targeting muscles even if patients cannot walk.

INTRODUCTION

Myotonic dystrophy type 1 (DM1) is an autosomal dominant inherited disorder related to the extension of a trinucleotide (CTG) repeat in the 3' -untranslated region of the myotonic dystrophy protein kinase gene. Although this expanded region is in a non-coding sequence, many symptoms arise from abnormal RNA [1]. Clinical features of DM1 include progressive muscle atrophy and weakness, grip and percussion myotonia, hatchet face, and involvement of the central nervous system, eyes, heart, and endocrine system. Expansion of the CTG repeats modifies clinical severity and age of onset. New therapeutic strategies have focused on abnormal RNA modification [2, 3].

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DM1 patients have metabolic syndromes including insulin resistance, increased body fat mass, and hypertriglyceridemia. A previous study reported decreased resting energy expenditure (REE) in DM1 patients [4]. REE is decreased in Duchene muscular dystrophy and Becker muscular dystrophy [5] but increased in Emery-Dreifuss muscular dystrophy [6]. It is difficult to measure total energy expenditure and basal energy expenditure, but REE may be measured by indirect calorimetry.

Body mass index is not useful for fat mass assessment in neuromuscular disorder patients because these patients have higher fat/muscle ratios due to fat infiltration into atrophied muscles. Dual energy X-ray absorptiometry is a noninvasive technique that provides regional estimations of fat-free mass, fat mass and bone mineral content [7, 8] and is useful for assessment in these patients.

This study aims to compare fat-free mass to fat mass in DM1 patients with matched controls and to evaluate the relationship of REE with whole and regional body composition in DM1 patients.

METHODS

Participants

Eighteen DM 1 patients aged 35 to 70 years (mean:48.89, sd:11.17) were recruited from our neurology department. DM1 was diagnosed by confirming the expansion of CTG repeats on genomic DNA extracted from peripheral blood leucocytes using Southern blot analysis in all patients. Disease severity was assessed using the muscular disability rating scale (MIRS) [9]. Computed tomography (CT) was performed in all patients to evaluate body muscle. We also studied blood cell counts, serum biochemistry and thyroid function.

Ten healthy volunteers were screened from our hospital co-worker as controls. They were matched with patients by age and gender.

Standard Protocols and Patient Consents

Ethic approval was obtained from the National Hospital Organization Asahikawa Medical Center Ethic Committee. All participants provided informed consent.

Measurements

REE was estimated by indirect calorimetry, using Fit 2200c (COSMED S.r.l., Rome, Italy). We measured at least one hour from the most recent meal.

Regional body composition was obtained by dual energy X-ray absorptiometry. All scans were performed and analyzed by a certified technician in our hospital. To measure regional body composition, the software divided the body into trunk (except for the head), left and right entire arms, and left and right entire legs. The whole body measurement was the sum of these components. The arm regions were delineated by a vertical line passing through the shoulder joint, and the leg regions were delineated by an oblique line passing through the femoral neck. The scans were analyzed with the enCORE software for body composition assessment: soft tissue mass; fat mass; fat-free mass; and bone mineral content. Body mass index was calculated by dividing body weight (kg) by height squared (m^2).

To estimate body muscle annually, all patients with DM 1 underwent CT scans. Thus, we estimated volumes of hypermetabolic organs from CT. REE for each of these organs was assessed using the specific resting metabolic rates (Ki, in kcal/kg per day) suggested by Elia et al.[10] for heart, liver, kidney, adipose tissue, and skeletal muscle. Skeletal muscle was calculated by subtracting total volume of heart, liver, and kidney from the fat-free mass measurement.

Statistical Analysis.

All analyses were performed using SPSS Version 20 (IBM) with a P-value of 0.05 as significant. Group comparisons between DM1 subjects and respective age-matched controls were performed using paired Student t-tests; chi-square tests were used for categorical variables. Within each group, we calculated Pearson correlation coefficients between REE and other variables, and performed multiple linear regression using variables that had P-values less than 0.20.

RESULTS

Demographics and Regional Body Composition.

Whole body and regional fat-free and fat masses were obtained from each participant. Demographic and anthropometric data are showed in Table 1. Body mass

index was greater than 25 kg/m2 in 4/18 of DM1 patients and 3/10 of controls. There was no significant difference between DM1 patients and controls for age, gender, height, weight, and body mass index.

Fat-free mass was lower in DM1 patients compared to controls (30.2 ± 5.93 kg vs. 44.8 ± 9.46 kg, P < 0.001).

Table 1. Demographics and anthropometric data.

		DM1 Patients	Controls	P-value
Age (year)		48.9 ± 11.2	49.9 ± 8.1	0.787
Female		7/18	4/10	0.954*
CTG repeat		1436 ± 447		
Height (m)		1.59 ± 0.01	1.63 ± 0.06	0.195
Weight (kg)		56.8 ± 8.2	64.2 ± 11.6	0.097
BMI		22.7 ± 3.8	24.1 ± 3.1	0.295
REE (kcal/day)		1178 ± 221	1560 ± 280	0.002
Left arm	Fat mass (kg)	0.874 ± 0.28	0.828 ± 0.33	0.725
	Fat-free mass (kg)	1.16 ± 0.38	2.40 ± 0.65	<0.001
Right arm	Fat mass (kg)	0.916 ± 0.28	0.873 ± 0.35	0.749
	fat-free mass (kg)	1.20 ± 0.34	2.53 ± 0.69	<0.001
Left leg	Fat mass (kg)	3.47 ± 1.0	2.86 ± 0.9	0.136
	Fat-free mass (kg)	4.11 ± 1.1	7.20 ± 1.5	<0.001
Right leg	Fat mass (kg)	3.39 ± 0.96	2.89 ± 0.95	0.207
	Fat-free mass (kg)	4.02 ± 1.1	7.30 ± 1.5	<0.001
Trunk	Fat mass (kg)	13.7 ± 4.6	10.5 ± 4.4	0.093
	Fat-free mass (kg)	16.7 ± 3.0	21.5 ± 4.5	0.002
Whole body	Fat mass (kg)	23.3 ± 6.8	18.7 ± 6.5	0.097
	Fat-free mass (kg)	30.2 ± 5.9	44.8 ± 9.4	<0.001

*All P-values based on paired t-tests except gender, which was calculated with a chi-square test

BMI: body mass index calculated as weight divided by height squared, CTG: trinucleotide, REE: resting energy expenditure.

There was also a trend for fat mass to be higher in DM1 patients (23.3 ± 6.84 kg vs. 18.7 ± 6.56 kg, P = 0.097).

General characteristics of DM1 patients

The mean CTG repeat expansion was 1426 (all patients' range: 400-2025) in DM1 patients. One patient (5.6%) was assigned MIRS = 3, four patients (22.2%) were MIRS = 4, and 13 patients (72.2%) were MIRS = 5. For all patients, we estimated the mean volumes of major organs from CT scans. Table 2 provides the mean estimated volumes of liver, heart, and kidneys, as well as adipose tissue and skeletal muscle. The results of blood tests in DM1 patients are shown in Table 3. One patient was diagnosed with hypothyroidism and treated by levothyroxine sodium hydrate.

REE

The REE was lower in DM1 patients than in controls (1178 ± 221 kcal/day vs. 1560 ± 280 kcal/day, P = 0.002). REEs from each organ were estimated in DM1 patients. We did not estimate from the brain (Table 2).

In controls, REE correlated with weight (r = 0.862; P = 0.001), height (r = 0.749; P = 0.006), body mass index (r = 0.775; P = 0.004), fat-free mass in trunk (r = 0.850; P = 0.001), fat-free mass in left arm (r = 0.737; P = 0.008), fat-free mass in right arm (r = 0.781; P = 0.004), fat-free mass in left leg (r = 0.808; P = 0.002), fat-free mass in right leg (r = 0.799; P = 0.003), and fat-free mass of the whole body (r = 0.837; P = 0.001). We further analyzed the relationship between REE and these variables using multiple linear regression. In the multiple linear regression analysis, REE of controls correlated only with weight (beta = 0.86; P = 0.001). The regression equation was REE = 20.81 ± weight (kg) + 224.

In DM1 patients, REE correlated with fat-free mass in all body composition (Table 1) measurements and some blood measurements (Table 3). We applied linear multiple regression using variables that had P < 0.20 for bivariate correlations. REE correlated with fat-free mass of whole body (beta = 0.805, P < 0.001) and gamma glutamyl transpeptidase (GGT) (beta = 0.352; P = 0.01) (Figure 1) with a regression equation of REE (DM1) = 30.0 ± WB FFM + 1.49 ± GGT + 140. We divided the DM1 patients into two groups based on severity (MIRS of 5 vs. lower MIRS). The REE was lower in patients with MIRS of 5

Table 2. Volume and REE of major organs in DM1 patients.

Organ	Volume	REE (kcal/day)
Heart (g)	364 ± 97	160 ± 42.7
Liver (g)	786 ± 244	157 ± 48.8
Kidney (g)	202 ± 56	88.9 ± 24.8
Skeletal muscle[a] (kg)	26.5 ± 5.40	344 ± 70.2
Adipose tissue[b] (kg)	23.3 ± 6.84	105 ± 30.8

Volumes estimated from CT. REE of each organ was obtained from volume and specific resting metabolic rates (Ki;K_{Heart}=440, K_{Liver}=200, K_{Kidney}=440, $K_{Skeletal\ muscle}$=13, $K_{Adipose\ tissue}$=4.5). [a]Skeletal muscle volume was obtained by subtracting volume of heart, liver and kidney from fat-free mass of whole body. [b]Adipose tissue was obtained by Dual X-ray Absorptiometry. REE: resting energy expenditure.

Table 3. Blood counts, serum biochemistry, and thyroid measures in DM1 patients.

Variable	Measurement	Reference Range, adults[a]
White-cell count (*10^4 er mm^3)	5.67 ± 1.87	3.50 – 9.00
Red-cell count (*10^6 per mm^3)	4.46 ±0.47	3.80 – 5.00
Hemoglobin (g/dl)	13.7 ± 1.63	11.1 - 15.1
Platelet count (*10^4 per mm^3)	16.6 ± 0.43	13.2 – 36.8
Total Protein (g/dl)	6.50 ± 0.55	6.7 - 8.3
Albumin (g/dl)	3.87 ± 0.44	4.0 - 5.0
Total bilirubin (mg/dl)	0.55 ± 0.25	0.3 - 1.2
Aspartate aminotransferase (U/l)	33 ± 12	13 -33
Alanine aminotransferase (U/l)	29 ± 13	6 – 27
Alkaline phosphatase (U/l)	295 ± 128	115 – 359
gamma-glutamyl transpeptidase (U/l)	88 ± 52	10 – 47
Urea nitrogen(mg/dl)	11.2 ± 3.5	8 – 22
Creatinine (mg/dl)	0.42 ± 0.15	0.4 - 0.7
Sodium (mmol/l)	145 ± 4.3	138 - 146
Potassium (mmol/l)	4.46 ± 0.67	3.6 - 4.9
Creatinephospho kinase (U/l)	167 ± 113	45 - 163
Hemoglobin A1c (%)	5.5 ± 0.6	4.6 - 6.2
Thyroid stimulating hormone (μU/l)	1.43 ± 1.0	0.35 - 3.73
Triiodothyronine (pg/ml)	2.44 ± 0.55	2.2 - 4.1
Thyroxine (ng/ml)	1.22 ± 0.24	0.88 - 1.81

[a]The reference ranges used at our hospital are for adults who do not have medical conditions that could affect the results.

Figure 1. Scatter plot between REE and FFM or GGT.

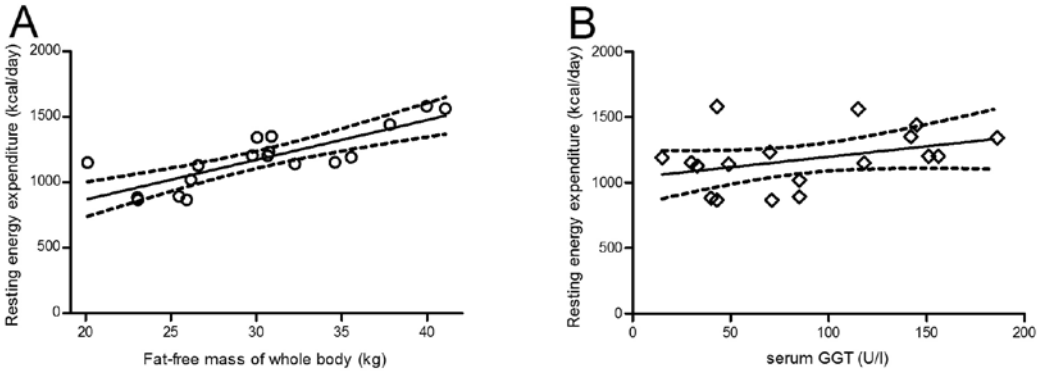

Figure 1. A: Scatter plot and correlation between REE and fat-free mass of whole body in DM1 patients. B: Scatter plot and correlation between REE and serum gamma glutamyl transpeptidase in DM1 patients. Line shows mean, dot line shows 95% Confidence interval. REE: resting energy expenditure.

(1095 ± 173.5 kcal/day vs. 1394 ± 192.9 kcal/day; P = 0.006).

DISCUSSION

We demonstrated that REE in DM1 patients was decreased compared with healthy controls. In addition, REE in DM1 patients correlated with fat-free mass of whole body and serum GGT.

REE in healthy controls correlated with body weight. We found that the difference between DM1 and controls in body composition was fat-free mass; there were no significant differences in fat mass in any region. Fat-free mass in this study is mainly composed from skeletal muscle and smooth muscle because bone mineral content was measured separately from other body composition in our dual energy X ray absorptiometry. We analyzed the REEs of major organs as predictor variables in multiple linear regressions. Finally these variables were excluded. This fact indicates that skeletal muscle plays an important role about REE.

Serum GGT is another predictor variable for REE in DM1 patients. Serum GGT reflects mainly liver disease. The elevated serum GGT in DM1 patients seems to be from nonalcoholic fatty liver disease. However, it is unclear that the relationship between fatty liver disease and REE. On the other hands, Generally elevation of serum GGT is associated with oxidant stress especially in liver cirrhosis patients[11] . Liver cirrhosis patients have higher REE than normal controls[12] . DM1 patients frequently have nonalcoholic fatty liver disease, which is associated with insulin resistance and features of metabolic syndromes[13] . Serum GGT is a marker of nonalcoholic fatty liver disease in patients with metabolic syndrome[14] and not only in DM1; it may also be elevated by a biliary system disorder such as gallstones. However, we could not detect gallstones in our DM1 patients in body muscle CT. This result indicates that liver dysfunction associated with REE in DM1 patients is similar to those of liver cirrhosis patients.

REE measured by indirect calorimetry is all metabolic energy that is not corrected from body profile. On the other hand, the Harris-Benedict equation is corrected from body profile. REE is more important than the Harris-Benedict equation in nutrition management of neuromuscular disease in abnormal body profile. We consider that body mass

index and other body region index are not useful in same reason.

This study has some limitations. First, the DM1 patients participated in this study are severely disabled. Seventeen patients had MIRS ratings greater than 4. Thereby we could not examine completely the relationship between REE and MIRS stages. Second, we estimated REE of major organs by evaluating their volumes with muscle CT. However, body muscle CT was originally performed to observe muscle atrophy and its distribution. It is difficult to measure the area of major organs exactly. We could not find a correlation between REE and major organs but one may exist. Third, healthy controls were recruited from our hospital. This may be a bias because hospital workers may have greater knowledge about health. This may result in more differences between the two groups.

In conclusion, we demonstrated the following two things. First, REE in DM1 patients was decreased relative to healthy controls. Second, REE in DM1 patients is proportional to the fat-free mass of whole body and gamma glutamyl transpeptidase. This fact indicates that REE in DM1 is associated with muscle volume and liver function. Thus, REE will be a good indicator of evaluating new therapies targeting muscles even if patients cannot walk.

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