

target Borg's scale ranged from 13 (fairly hard) to 15 (hard). Subjects measured their heart rates by palpation while walking and jogging, and recorded the duration (min) and intensity (heart rate or the Borg's scale) during each exercise session.

Statistical analysis

Values are expressed as means ± S.D. Unpaired *t*-tests were used to test difference in changes in variables between the two treatment groups. Paired *t*-tests were used to assess differences between variables before and after the weight reduction programs. Relationships between two measurement variables were assessed by Pearson's product moment correlation. Multiple regression analyses with the forward stepwise method were performed to estimate the independent contribution of the selected variables to the variations in fasting tHcys at baseline and changes in fasting tHcys concentration in response to weight reduction. For tHcys concentration and $\dot{V}O_{2max}$, a two (group) by two-way (time) ANOVA was performed to determine group-by-time interaction.

Fisher's post hoc test for multiple comparisons with Bonferroni corrections were performed if the ANOVA was significant. According to a statement of the American Heart Association [19,20], we classified subjects into two groups: 'Normal' group with tHcys <15 μmol/L and 'High (hyperhomocysteinemia)' group with tHcys ≥15 μmol/L. Analyses were performed on log-transformed values for the variables that were not normally distributed. Probability values below 0.05 were regarded as significant. The data were analyzed with the SPSS (Chicago, IL), version 11.0J for Windows.

Results

Attendance at the exercise training (40 sessions) averaged 80% (range 37.5–100%) for the subjects from the DE group. The frequency of the exercise training was 2.4 ± 0.6 day/weeks with an average duration of 95 ± 27 min/weeks. The average heart rates during walking and jogging were 122 ± 17 beats/min and 141 ± 18 beats/min,

Table 2 Baseline data and changes in measurement variables for groups with diet alone and diet plus exercise

	Diet alone		Diet plus exercise	
	Baseline (n = 24)	Change (n = 21)	Baseline (n = 24)	Change (n = 22)
Body weight (kg)	78.3 ± 5.9	-7.8 ± 3.2***	79.5 ± 7.1	-9.1 ± 3.6***
Body mass index (kg/m ²)	27.8 ± 1.7	-2.7 ± 1.1***	27.6 ± 1.7	-3.1 ± 1.1***
Waist circumference (cm)	94.9 ± 11.0	-9.5 ± 3.5***	95.6 ± 10.2	-8.4 ± 3.7***
Hip circumference (cm)	98.7 ± 3.6	-4.3 ± 2.0***	100.7 ± 6.8	-5.0 ± 2.3***
Percentage fat mass (%)	30.7 ± 3.7	-6.8 ± 3.1***	30.6 ± 4.6	-8.2 ± 3.6***
Fat mass (kg)	24.1 ± 3.9	-7.1 ± 2.7***	26.5 ± 8.5	-8.7 ± 3.1***
Fat free mass (kg)	54.1 ± 4.3	-0.7 ± 1.3	57.0 ± 8.5	-0.5 ± 1.6
Visceral fat area (cm ²)	171 ± 60	-41 ± 42***	172 ± 60	-60 ± 27***
Subcutaneous fat area (cm ²)	186 ± 57	-45 ± 26***	213 ± 111	-53 ± 23***
Total fat area (cm ²)	357 ± 91	-87 ± 62***	385 ± 155	-113 ± 41***
Systolic blood pressure (mmHg)	129 ± 17	-9 ± 12**	135 ± 21	-13 ± 15***
Diastolic blood pressure (mmHg)	87 ± 13	-9 ± 8**	90 ± 11	-12 ± 10**
Homocysteine (μmol/L)	14.2 ± 2.5	-0.3 ± 2.7	14.4 ± 5.8	-2.3 ± 5.0
Folate (nmol/L)	6.1 ± 1.6	0.1 ± 0.0	6.1 ± 2.0	0.6 ± 1.7
Vitamin B ₆ (nmol/L)	15.6 ± 7.3	-2.5 ± 6.9	16.6 ± 10.6	-0.7 ± 6.2
Vitamin B ₁₂ (nmol/L)	434 ± 168	-5 ± 82	456 ± 133	7 ± 120
TC (mg/dL)	231 ± 37	-31 ± 22***	213 ± 37	-31 ± 35***
HDLc (mg/dL)	53 ± 10	2 ± 9	53 ± 13	4 ± 10
LDLc (mg/dL)	141 ± 36	-21 ± 27**	126 ± 36	-17 ± 29**
HDLc/TC	0.24 ± 0.05	0.05 ± 0.04***	0.26 ± 0.09	0.06 ± 0.08**
Triglycerides (mg/dL)	180 ± 139	-58 ± 70**	169 ± 93	-95 ± 88***
Fasting glucose (mg/dL)	101 ± 14	-4 ± 13	106 ± 23	-12 ± 18**
Insulin (μU/mL)	11.3 ± 2.7	-0.9 ± 6.1	13.4 ± 4.4	-5.1 ± 4.9***
HOMA-R	2.80 ± 0.86	-0.32 ± 1.20	3.49 ± 1.43	-1.50 ± 1.81**
Maximal oxygen intake (L/min)	2.49 ± 0.57	-0.07 ± 0.22	2.57 ± 0.48	0.05 ± 0.36

Data are expressed as mean ± S.D. TC, total cholesterol; HDLc, high-density lipoprotein cholesterol; LDLc, low-density lipoprotein cholesterol; HOMA-R = homeostasis model assessment. NS = non-significance, *p < 0.05, **p < 0.01, ***p < 0.001 within difference (pre vs. post).

respectively, which corresponded to $52 \pm 15\%$ and $76 \pm 17\%$ of $\dot{V}O_{2\max}$. The mean energy expenditure during the exercise training was estimated for approximately 500 kcal/weeks. Number of subjects in current cigarettes smoking was eight (17%), consisting of four in the DA and four in the DE. Of them, one subject smokes for 10 cigarettes per day, five for 20, one for 25, and one for 40.

No difference was found in any variables between groups with DA and DE at baseline (Table 2). For both groups body weight and other measurements in CT, DXA and anthropometric variables decreased significantly. Concentrations of tHcy, folate, vitamin B₆ and B₁₂ remained unchanged for the DA group. A significant decrease was found in tHcys concentration of the DE group. The repeated ANOVA revealed that there was non-significant group-by-time interaction in tHcys concentration and $\dot{V}O_{2\max}$. For the DE group $\dot{V}O_{2\max}$ tended to increase ($p=0.09$) from 2.57 ± 0.48 to 2.63 ± 0.48 mL/min. On the other hand, for the DA group $\dot{V}O_{2\max}$ tended to decrease ($p=0.07$) from 2.49 ± 0.57 to 2.42 ± 0.56 mL/min.

We quantified the independent contributions of the variables considered here to the variance in tHcys concentration (Table 3). At baseline, insulin ($p < 0.001$) was the best predictor of tHcys concentration. Number of current cigarettes per day ($p=0.005$) and vitamin B₆ ($p=0.009$) were also significant predictors of tHcys concentration. When change in tHcys concentration was defined as the dependent variable, changes in insulin ($p=0.032$) and $\dot{V}O_{2\max}$ ($p=0.048$) were selected as significant predictors.

Fig. 1 displays individual tHcys data for the 'High' ($n=17$) and 'Normal' ($n=27$) groups at

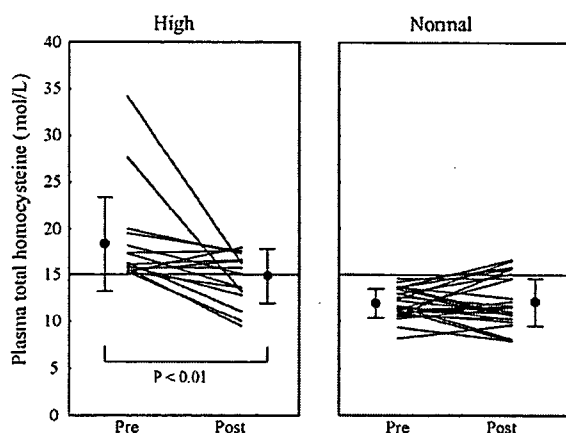


Figure 1 Plasma total homocysteine concentration ($\mu\text{mol/L}$) in the 'High' and 'Normal' groups at baseline and post weight reduction. Vertical lines indicate means (●) and S.D.

Table 3 Results of multiple regression analysis

Baseline homocysteine ($n=48$)		Change in homocysteine ($n=43$)	
Independent variable	Beta	P	Model R ²
Insulin	0.338	<0.001	26.7
Number of current cigarettes per day	0.165	0.0005	39.7
Vitamin B ₆	-0.148	0.009	48.2
Independent variable	Beta	P	Partial R ²
Δ Insulin	0.664	0.032	13.4
Δ Maximal oxygen intake	-3.78	0.048	7.2
			Model R ²
			13.4
			20.6

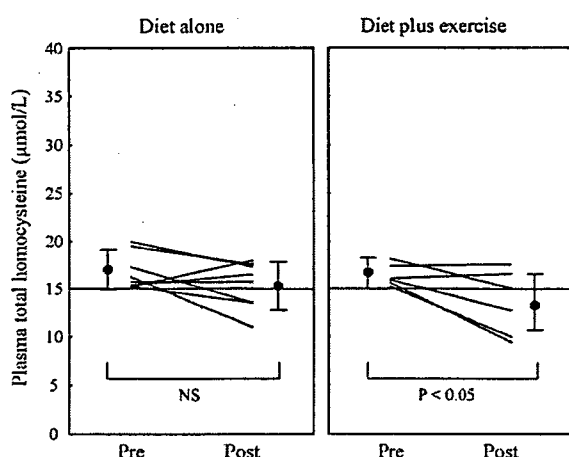


Figure 2 Plasma total homocysteine concentration ($\mu\text{mol/L}$) of the groups with diet alone and diet plus exercise in the 'High' group at baseline and post weight reduction. Vertical lines indicate means (\bullet) and S.D.

baseline and after weight reduction. Mean values of tHcys concentration decreased from 18.3 ± 5.1 to $14.7 \pm 2.7 \mu\text{mol/L}$ ($p < 0.01$) for the 'High' group but did not change for the 'Normal' group ($11.9 \pm 1.5 \mu\text{mol/L} \rightarrow 12.0 \pm 2.6 \mu\text{mol/L}$). Significant difference was found in the tHcys change between the two groups. After weight reduction, 11 (65%) individuals of the 'High' group decreased their tHcys. Seven of the 11 individuals decreased their tHcys concentrations to less than $15 \mu\text{mol/L}$. The tHcys concentrations of 21 (78%) of subjects in the 'Normal' group remained unchanged under $15 \mu\text{mol/L}$.

For both 'High' and 'Normal' groups, Folate ($5.9 \pm 2.1 \text{ nmol/L} \rightarrow 5.9 \pm 2.4 \text{ nmol/L}$ and $6.1 \pm 1.5 \text{ nmol/L} \rightarrow 6.6 \pm 2.2 \text{ nmol/L}$, respectively), vitamin B₆ ($14.8 \pm 8.4 \text{ nmol/L} \rightarrow 12.1 \pm 4.6 \text{ nmol/L}$ and $15.7 \pm 5.2 \text{ nmol/L} \rightarrow 14.9 \pm 4.7 \text{ nmol/L}$, respectively) and B₁₂ ($429 \pm 166 \text{ nmol/L} \rightarrow 422 \pm 153 \text{ nmol/L}$ and $459 \pm 140 \text{ nmol/L} \rightarrow 467 \pm 155 \text{ nmol/L}$, respectively) remained unchanged between baseline and after weight reduction.

For the 'High' group, mean values of tHcys concentration decreased from 20.1 ± 7.0 to $13.9 \pm 3.0 \mu\text{mol/L}$ ($p < 0.01$) in the DE group ($n = 8$) but did not change in the DA group ($16.6 \pm 1.9 \mu\text{mol/L} \rightarrow 15.4 \pm 2.3 \mu\text{mol/L}$, $n = 9$). Furthermore, to consider a statistical phenomenon "regression to the mean" [21], we excluded two subjects with the highest two tHcys values ($34.2 \mu\text{mol/L}$ and $27.6 \mu\text{mol/L}$) of the DE group. The DE group ($n = 6$) still displayed a significant reduction ($16.5 \pm 1.1 \mu\text{mol/L} \rightarrow 13.6 \pm 3.4 \mu\text{mol/L}$, $p < 0.05$) after the exclusion (Fig. 2).

Discussion

We have examined the effect of weight reduction on tHcys concentration in obese Japanese men, and tested whether adding aerobic exercise training (a brisk walking and a mild jogging) to a dietary weight-reduction program would further reduce tHcys concentration over diet alone. This is the first study to examine whether weight reduction can be a factor for reducing tHcys in obese individuals. Our main observation is that the 14-week weight-reduction program with aerobic exercise training has reduced significantly tHcys concentration. Boushey et al. [4] have reported that a total of 10% of the population's coronary artery risk appears attributable to homocysteine, and an increase of $5 \mu\text{mol/L}$ in tHcys raises the risk of coronary artery disease by 60% in men and 80% in women. Schnyder et al. [5] found that lowering tHcys levels from 11.1 to $7.2 \mu\text{mol/L}$ reduced significantly the rate of coronary restenosis after angioplasty and decreased the incidence of major adverse cardiac events. In a more recent study, lowering tHcys level by 25% ($3 \mu\text{mol/L}$) was associated with an 11% lower ischemic heart disease and 19% lower stroke risk [6]. Therefore, our observation of lowering tHcys concentration in the DE group may contribute to reduce the risk of serious heart events, especially for individuals with hyperhomocysteinemia.

Little information is available on the effect of exercise on tHcy level. A few epidemiological studies have reported that high level of daily physical activity was associated with low tHcys concentration [22,23], although the precise mechanisms on the association between tHcys concentration and physical activity or maximal oxygen uptake are still unclear. Some studies have reported that a short-term (less than 8 weeks) regular exercise did not affect tHcys levels [24,25]. A study by Randeve et al. [12] reported that women performed, on average, 23.8 ± 5.0 min of brisk walking per day, at least 3 walks per week for 6 months, and their $\dot{V}O_{2\text{max}}$ increased significantly from 2.50 ± 0.31 to $2.60 \pm 0.35 \text{ L/min}$, which resulted in a significant reduction in their tHcys (from 10.1 ± 3.2 to $7.4 \pm 2.0 \mu\text{mol/L}$). Our data suggests that an increase in $\dot{V}O_{2\text{max}}$ associated with a reduction in tHcys concentration. The amounts of exercise training, calculated as products of exercise duration (min) \times frequency (times/weeks), were rather greater for our subjects (95 min/weeks) than for the women of the above study ($23.8 \times 3 = 71.4$ min/weeks) by Randeve et al. [12]. Body fat change is well-known to affect cardiorespiratory fitness ($\dot{V}O_{2\text{max}}$). Our data displayed

that changes in fat mass and fat area had a trend to be associated with a change in homocysteine level, but that's not significant against our expectations.

It is well recognized that insulin decreases plasma methionine. The insulin-induced fall in plasma methionine concentration may be mediated through increased tissue uptake of methionine or metabolism via the transsulfuration pathway resulting in increased tHcys concentration. In addition, Solini et al. [26] reported that folic acid (folate) supplementation reduced plasma tHcys concentration and ameliorated insulin sensitivity. A recent study [27] has demonstrated an inverse relationship between tHcys concentration and insulin sensitivity in women with preeclampsia. Our data also indicates that insulin is one of the significant predictors for tHcys concentration. However, because of the multiple abnormalities in metabolism and endothelial function present in preeclampsia, it is not clear whether a cause-effect relationship exists between the two variables [1].

Folic acid and vitamin B₁₂ regulate metabolic pathways catalyzed by the enzymes methylenetetrahydrofolate reductase (MTHFR) and methionine synthase, respectively, whereas pyridoxine (vitamin B₆) is a cofactor for cystathionine beta synthase on the transsulfuration pathway [19]. A number of studies have shown that inverse relationships of tHcys concentration with plasma/serum levels of folate, vitamin B₆ and vitamin B₁₂ [28,29]. Our data indicates that vitamin B₆ is one of the significant predictors for tHcys concentration. A recent study by Heart Outcomes Prevention Evaluation (HOPE) 2 Investigators [30] however, demonstrated that supplementation combining folic acid and vitamins B₆ and B₁₂ did not reduce the risk of cardiovascular events in patients with vascular disease. On the other hand, according to our observation, the diet plus exercise training contributed to not only lower tHcys concentrations but also reduce body weight and visceral fat area which are well-known to be risk factors for vascular disease.

Several studies have reported a positive association between cigarette smoking and tHcys concentration [23,31–33]. Although the exact mechanism by which cigarette smoking increases tHcys is not known, the association between smoking and tHcys can be explained by low concentrations of blood folate, vitamin B₆, and vitamin B₁₂ in smokers [31]. In the current study, current number of cigarettes per day and Brinkman index (current number of cigarettes per day × years) were used as candidate predictors for tHcys level, but

Brinkman index was not selected. These results suggest that current smoking status may be more associated with tHcys level.

A limitation of this study is that subjects' actual dietary energy intakes and essential nutrients (i.e., minerals and vitamins) were not controlled precisely. The precise control of diet was difficult in this study employing free living subjects. Several vitamins function as cofactors and substrates in the metabolism of methionine and homocysteine. As we mentioned above, studies have shown the inverse relationships of tHcys concentration with plasma/serum levels of folate, vitamin B₆, and vitamin B₁₂ [29,34]. It is possible that a difference in intakes of folic acid and vitamins B₆ and B₁₂ between the groups would induce a variability of plasma levels of folate, vitamin B₆, and vitamin B₁₂, which may partly preclude our definitive conclusions. In our data, however, there was a non-significant change in plasma levels of folate, vitamin B₆, and vitamin B₁₂ within each treatment group, and no difference was found in the changes between the DA and DE groups. These results documented that the change in intakes of folic acid and vitamins B₆ and B₁₂ would be little in all subjects.

In conclusion, our data raises the possibility that for obese men, tHcys concentration is lowered with diet plus aerobic exercise training although weight reduction with diet alone did not improve tHcys levels. Adding aerobic exercise training to a dietary weight-reduction program is necessary for a reduction in tHcys.

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References

- [1] Fonseca V, Guba SC, Fink LM. Hyperhomocysteinemia and the endocrine system: implications for atherosclerosis and thrombosis. *Endocr Rev* 1999;20:738–59.
- [2] Langman LJ, Ray JG, Evrovski J, Yeo E, Cole DE. Hyperhomocyst(e)inemia and the increased risk of venous thromboembolism: more evidence from a case-control study. *Arch Intern Med* 2000;160:961–4.

- [3] Tanne D, Haim M, Goldbourt U, Boyko V, Doolman R, Adler Y, et al. Prospective study of serum homocysteine and risk of ischemic stroke among patients with preexisting coronary heart disease. *Stroke* 2003;34:632–6.
- [4] Boushey CJ, Beresford SA, Omenn GS, Motulsky AG. A quantitative assessment of plasma homocysteine as a risk factor for vascular disease. Probable benefits of increasing folic acid intakes. *JAMA* 1995;274:1049–57.
- [5] Schnyder G, Roffi M, Pin R, Flammer Y, Lange H, Eberli FR, et al. Decreased rate of coronary restenosis after lowering of plasma homocysteine levels. *N Engl J Med* 2001;345:1593–600.
- [6] Homocysteine Studies Collaboration. Homocysteine and risk of ischemic heart disease and stroke: a meta-analysis. *JAMA* 2002;288:2015–22.
- [7] Kenchaiah S, Evans JC, Levy D, Wilson PW, Benjamin EJ, Larson MG, et al. Obesity and the risk of heart failure. *N Engl J Med* 2002;347:305–13.
- [8] Mokdad AH, Ford ES, Bowman BA, Dietz WH, Vinicor F, Bales VS, et al. Prevalence of obesity, diabetes, and obesity-related health risk factors, 2001. *JAMA* 2003;289:76–9.
- [9] Vague P. The degree of masculine differentiation of obesity. *Am J Clin Nutr* 1956;4:20–34.
- [10] Rasmussen LB, Ovesen L, Bülow I, Knudsen N, Laurberg P, Perrild H. Folate intake, lifestyle factors, and homocysteine concentrations in younger and older women. *Am J Clin Nutr* 2000;72:1156–63.
- [11] Folsom AR, Nieto FJ, McGovern PG, Tsai MY, Malinow MR, Eckfeldt JH, et al. Prospective study of coronary heart disease incidence in relation to fasting total homocysteine, related genetic polymorphisms, and B Vitamins: the atherosclerosis risk in communities (ARIC) study. *Circulation* 1998;98:204–10.
- [12] Randevo HS, Lewandowski KC, Drzewoski J, Brooke-Wavell K, O'Callaghan C, Czupryniak L, et al. Exercise decreases plasma total homocysteine in overweight young women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 2002;87:4496–501.
- [13] The Examination Committee of Criteria for 'Obesity Disease' in Japan, Japan Society for the Study of Obesity. New criteria for 'obesity disease' in Japan. *Circ J* 2002;66:987–92.
- [14] Durand P, Fortin LJ, Lussier-Cacan S, Davignon J, Blache D. Hyperhomocysteinemia induced by folic acid deficiency and methionine load—applications of a modified HPLC method. *Clin Chim Acta* 1996;252:83–93.
- [15] Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972;18:499–502.
- [16] Brinkman GL. The effect of bronchitis, smoking and occupation on ventilation. *Am Rev Respir Dis* 1963;87:684–93.
- [17] Okura T, Nakata Y, Yamabuki K, Tanaka K. Regional body composition changes exhibit opposing effects on coronary heart disease risk factors. *Arterioscler Thromb Vasc Biol* 2004;24:923–9.
- [18] Borg G. Perceived exertion: a note on "history" and methods. *Med Sci Sports* 1973;5:90–3.
- [19] Kang SS, Wong PW, Malinow MR. Hyperhomocyst(e)inemia as a risk factor for occlusive vascular disease. *Annu Rev Nutr* 1992;12:279–98.
- [20] Malinow MR, Bostom AG, Krauss RM. Homocyst(e)ine, diet, and cardiovascular diseases: a statement for healthcare professionals from the Nutrition Committee, American Heart Association. *Circulation* 1999;99:178–82.
- [21] Bland JM, Altman DG. Regression towards the mean. *BMJ* 1994;308:1499.
- [22] Mennen LJ, de Courcy GP, Guillard JC, Ducros V, Bertrais S, Nicolas JP, et al. Homocysteine, cardiovascular disease risk factors, and habitual diet in the French Supplementation with Antioxidant Vitamins and Minerals Study. *Am J Clin Nutr* 2002;76:1279–89.
- [23] Nygard O, Vollset SE, Refsum H, Stensvold I, Tverdal A, Nordrehaug JE, et al. Total plasma homocysteine and cardiovascular risk profile. The Hordaland Homocysteine Study. *JAMA* 1995;274:1526–33.
- [24] De Cree C, Malinow MR, van Kranenburg GP, Geurten PG, Longford NT, Keizer HA. Influence of exercise and menstrual cycle phase on plasma homocysteine levels in young women—a prospective study. *Scand J Med Sci Sports* 1999;9:272–8.
- [25] Volek JS, Gomez AL, Love DM, Weyers AM, Hesslink Jr R, Wise JA, et al. Effects of an 8-week weight-loss program on cardiovascular disease risk factors and regional body composition. *Eur J Clin Nutr* 2002;56:585–92.
- [26] Solini A, Santini E, Ferrannini E. Effect of short-term folic acid supplementation on insulin sensitivity and inflammatory markers in overweight subjects. *Int J Obes* 2006;30:1197–202.
- [27] Laivuori H, Kaaja R, Turpeinen U, Viinikka L, Ylikorkala O. Plasma homocysteine levels elevated and inversely related to insulin sensitivity in preeclampsia. *Obstet Gynecol* 1999;93:489–93.
- [28] Robinson K, Arheart K, Refsum H, Brattstrom L, Boers G, Ueland P, et al. Low circulating folate and vitamin B₆ concentrations: risk factors for stroke, peripheral vascular disease, and coronary artery disease: European COMAC Group. *Circulation* 1998;97:437–43.
- [29] Selhub J, Jacques PF, Wilson PW, Rush D, Rosenberg IH. Vitamin status and intake as primary determinants of homocysteinemia in an elderly population. *JAMA* 1993;270:2693–8.
- [30] Heart Outcomes Prevention Evaluation (HOPE) 2 Investigators. Homocysteine lowering with folic acid and B vitamins in vascular disease. *N Engl J Med* 2006;354:1567–77.
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- [31] Ganji V, Kafai MR. Demographic, health, lifestyle, and blood vitamin determinants of serum total homocysteine in the third National Health and Nutrition Examination Survey, 1988–1994. *Am J Clin Nutr* 2003;77:826–33.
- [32] Jacques PF, Bostom AG, Wilson PWF, Rich S, Rosenberg IH, Selhub J. Determinants of plasma total homocysteine con-

- centration in the Framingham Offspring cohort. *Am J Clin Nutr* 2001;73:613–21.
- [33] de Bree A, Verschuren WM, Blom HJ, Kromhout D. Lifestyle factors and plasma homocysteine concentrations in a general population sample. *Am J Epidemiol* 2001;154:150–4.
- [34] Kang SS, Wong PWK, Norusis M. Homocysteinemia due to folate deficiency. *Metabolism* 1987;36:458–62.

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The influence of physical activity-induced energy expenditure on the variance in body weight change among individuals during a diet intervention

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KEYWORDS

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Summary

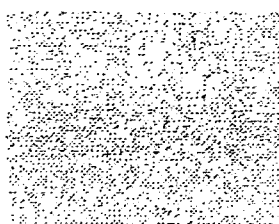
Objective: We investigated the relationship between the variability in body weight change among individuals and diet restriction or physical activity during a 14-week intervention.

Design: A prospective clinical trial with a 14-week weight reduction intervention design. In total, 90 middle aged, Japanese, obese women enrolled as subjects.

Measurements: The outcome variable was the change in body weight during the intervention period. Other primary variables were total energy intake, carbohydrate intake, fat intake, protein intake, total energy expenditure (TEE), and activity energy expenditure (AEE). Diet intake was assessed by 3 days, weighed dietary records and dietary recall interviews. Physical activity was assessed by a uniaxial accelerometry sensor and a diary of exercise.

Results: Significant reductions were observed in body weight (−8.5 kg) as a result of intervention. When the subjects were assigned to three categories depending on AEE during intervention, the loss of body weight was significantly greater for subjects within the *upper* (−9.6 kg) AEE category than for those in the *middle* (−8.5 kg) or *lower* AEE (−7.5 kg) categories. In addition, a significant correlation ($r = 0.57$, $p < 0.0001$) was observed between a subject's AEE before and during the intervention. On the other hand, no significant correlation was observed between

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body weight reduction and energy intake, indicating that strict diet restriction does not always result in a large weight loss.

Conclusion: Activity energy expenditure, not only through voluntary exercise but also through spontaneous, daily, physical activities can have a positive effect on reducing body weight.

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Introduction

Obesity is closely associated with some major lifestyle-related disorders such as hypertension, dyslipidemia, type 2 diabetes, and cardiovascular disease [1,2]. In addition, the prevalence of obesity continues to increase in many countries [3]. In Japan, despite the fact that the Japanese population has only a 2–3% "obesity category" [4], defined by the World Health Organization as a body mass index (BMI) ≥ 30 kg/m², the prevalence of lifestyle-related disorders are high ([5,6]). This suggests that, even with mild obesity, Japanese individuals are likely to develop lifestyle-related disorders. Therefore, the Japan Society for the Study of Obesity (JASSO) originally defined obesity as a BMI ≥ 25 kg/m² [7]. On the other hand, it is well known that many of the risk factors related to obesity tend to improve after body weight reduction. Some studies [8–11] have indicated that coronary heart disease (CHD) risk factors such as intra-abdominal fat area, blood pressure, and biochemical variables (triglycerides, fasting plasma glucose, etc.) improve in response to weight reduction achieved through a strictly supervised weight reduction intervention.

However, although obese individuals may participate in the same intervention program for weight reduction, changes in the values of CHD risk factors tend to differ among individuals; there is a lot of variability in the mean values of the CHD risk factors [9–11]. Changes in these risk factors are known to be closely associated with the change in body weight [10,12]. The variances in CHD risk factors could be due to the difference in body weight reduction among individuals even when they participate in the same weight-reduction program. Therefore, if weight management instructors take into consideration the variability in body weight change among individuals, they could provide more effective prescriptions regarding physical activity and diet for obese individuals.

In general, the most important factor for reducing body weight is a balance between energy intake and energy expenditure [13]. Instructors of body

weight reduction programs should indicate the proper diet and appropriate physical activity for their patients. Understandably, they cannot control everything in a participant's daily life, and as a result, it is inevitable that the change in energy balance would vary among individuals. The body weight might decrease resulting when energy expenditure exceeds energy intake. Diet restriction and increased physical activity could have a strong effect on creating such imbalances.

The effect of diet restriction and physical activity on changes in body weight may be the most important considerations when investigating the variance in body weight change among individuals. Actually, little is known about the relationship between variable body weight changes among individuals and diet restriction or physical activity.

Taking these factors into consideration, in this study we examined whether the variance in body weight change among individuals may be influenced by energy intake (diet restriction) or energy expenditure (physical activity).

Methods

Subjects

Subjects were recruited through advertisements in local newspapers, and 111 women were initially screened. We included individuals for our study who met at least one of following two criteria: (1) BMI > 25 kg/m²; or (2) intra-abdominal fat area at the level of the umbilicus > 100 cm², as determined by CT scan. None of the subjects had any cardiac disease. We also excluded individuals who were unable to perform all measurements for personal reasons. In total, 90 women, aged 30–65 years, with a baseline mean BMI of 27.8 kg/m² (22.5–34.6 kg/m²) (Table 1) were selected for the study. The aim and design of the study were explained to each subject before they gave their written informed consent. This study was approved by the Ethical Committee of the University of Tsukuba.

Table 1 Participants' descriptive characteristics and any changes in values during the intervention period

	Week 0	Week 10	Week 15	d
n = 90				
Age (Year)	52.3 ± 7.1 (30-65)			
Height (cm)	156.3 ± 4.9 (144.1-167.9)			
Body weight (kg)	68.1 ± 8.4 (52.0-92.6)	62.9 ± 7.4 (49-86)	59.5 ± 7.6 (46.6-83.6)	-8.5 ± 2.4*
Body mass index (kg/m ²)	27.8 ± 2.6 (22.5-34.6)	25.8 ± 2.3 (21.3-32.4)	24.4 ± 2.4 (20.1-31.8)	-3.4 ± 0.9**
Waist circumference (cm)	95.5 ± 8.1 (80.5-120.9)		87.1 ± 7.6 (71.4-114.3)	-8.4 ± 3.6**
Intra-abdominal fat area (cm ²)	114.6 ± 40.6 (39.3-225.3)		84.9 ± 34.4 (27.1-188.1)	-30 ± 24*
Systolic blood pressure (mmHg)	132 ± 19 (96-218)		117 ± 16 (90-158)	-15 ± 13*
Diastolic blood pressure (mmHg)	90 ± 10 (64-113)		73 ± 10 (54-98)	-9 ± 8*
Total cholesterol (mg/dl)	226 ± 40 (130-367)		199 ± 33 (121-304)	-27 ± 26**
Triglycerides (mg/dl)	121 ± 92 (38-680)		74 ± 30 (31-183)	-47 ± 79**
High-density lipoprotein cholesterol (mg/dl)	60 ± 13 (35-89)		61 ± 12 (40-92)	1 ± 8
Low-density lipoprotein cholesterol (mg/dl)	142 ± 36 (75-282)		123 ± 30 (57-235)	-19 ± 25**
Fasting plasma glucose (mg/dl)	100 ± 20 (77-210)		91 ± 9 (73-120)	-9 ± 16**

Values are presented as the mean ± standard deviation and ranges. Week 0, before weight reduction program; week 10, at the beginning of the 10th week of the weight reduction program; week 15, after weight reduction program; d: (15 wk) - (0 wk).
*p < 0.05; **p < 0.01.

Anthropometric variables

Body weight was measured to the nearest 0.1 kg using a digital scale (TBF-215; Tanita, Tokyo, Japan), height was measured to the nearest 0.1 cm using a wall-mounted stadiometer (TBF-215; Tanita, Tokyo, Japan), and BMI was calculated as weight (in kilograms) divided by height squared (meters squared). Waist circumference was measured to the nearest 0.1 cm at the level of the umbilicus with subjects in the standing position.

Abdominal adipose tissue area by CT

Intra-abdominal fat area (IFA) (cm²) was measured at the level of the umbilicus (L4-L5) using CT scans (TSX-002A; Toshiba, Tokyo, Japan) performed on subjects in the supine position. The IFA was calculated using a computer software program (FatScan; N2 system, Osaka, Japan).

Blood pressure and biochemical assays of blood

Systolic and diastolic blood pressures (SBP and DBP) were taken from the right arm using a mercury manometer after the subjects rested at least 20 min in a sitting position. Cuff sizes were selected based on upper arm girth and length.

A blood sample of ~10 ml was drawn from each subject after an overnight fast. Serum total cholesterol (TC) and triglycerides (TG) were determined enzymatically, serum high-density lipoprotein-cholesterol (HDL-C) was measured by the heparin-manganese precipitation method and fasting plasma glucose (FPG) was assayed by a glucose oxidase method. Serum low-density lipoprotein-cholesterol (LDL-C) was estimated according to the equation of Friedewald et al. [14]: LDL-C = total cholesterol - (HDL-C + triglycerides/5).

Dietary assessments

Total energy intake (in kilocalories) and the amounts of each nutrient (carbohydrates, fat, and protein in grams) before and at the beginning of week 10 of the weight reduction program were assessed by both 3-day weighed dietary records (3-day WDR) and dietary recall interviews for each subject performed by one skilled dietician. The dietician explained how to complete the 3-day WDR in detail before the measurements. The subjects learned how to use a digital cooking scale for weighing food. The dietician collected the recorded sheets from the 3-day WDR and codified the food items and food weights. Thereafter, the dietician

Table 2 Changes in energy intake and energy expenditure

n = 90	Week 0	Week 10	d
Total energy intake (kcal/d)	1867 ± 332 (1292–2816)	1195 ± 127 (943–1545)	-671 ± 325*
Carbohydrates intake (g/d)	259 ± 50 (119–410)	169 ± 24 (114–237)	-90 ± 49*
Fat intake (g/d)	57 ± 16 (30–97)	32 ± 8 (17–63)	-25 ± 17*
Protein intake (g/d)	70 ± 12 (41–101)	60 ± 9 (37–81)	-10 ± 14*
Total energy expenditure (kcal/d)	1916 ± 187 (1558–2365)	1855 ± 165 (1488–2262)	-61 ± 112*
Activity energy expenditure (kcal/d)	455 ± 111 (236–755)	474 ± 105 (246–823)	20 ± 100†
Total energy expenditure/body weight (kcal/kg d)	28.3 ± 2.2 (23.0–34.4)	29.7 ± 2.5 (23.9–37.1)	1.4 ± 1.8*
Activity energy expenditure/body weight (kcal/kg d)	6.7 ± 1.6 (3.7–11.1)	7.6 ± 1.8 (3.9–13.9)	0.9 ± 1.6*

Values are presented as the mean ± standard deviation and ranges. Week 0, before weight reduction program; week 10, at the beginning of the 10th week of the weight reduction program; d, (10wk) - (0wk).

* $p < 0.01$.

† $p < 0.1$.

interviewed each subject to elicit more information about the subject's food intake for the 3-day period. Based on the Standard Tables of Food Composition in Japan fourth revised edition [15], total energy intake along with carbohydrate, fat, and protein intakes were calculated (Table 2).

Energy expenditure assessments

The total energy expenditure (TEE) and activity energy expenditure (AEE) were assessed by both a uniaxial accelerometry sensor (Lifecorder; Suzuken Co. Ltd.) and a diary of exercise. Lifecorder can assess two types of AEE by activity level: energy expenditure of activities (EE_{Act}) and energy expenditure of minor activities ($EE_{minorAct}$) [16]. In this study, AEE was estimated as a total of these two EEs ($AEE = EE_{Act} + EE_{minorAct}$). Subjects wore the Lifecorder constantly (except while sleeping or bathing), for a 7-day period prior to the weight reduction program and for 7 days during week 10 of the weight reduction program. During these 7-day segments, the subjects were also instructed to keep a diary of their exercise. The diary consisted of the status of wearing the Lifecorder and detailed exercise information (i.e. type, time, and ratings of perceived exertion (RPE) [17]). For times of exceptional physical exertion (e.g. swimming), we estimated this exceptional EE by its metabolic equivalent (MET) from the diary entries. For these measurement periods, we calculated TEE and AEE from both the Lifecorder data and the diary entries.

Subjects were classified into the following three categories with regard to their AEE during the intervention: *upper*, the top 25% of participants in terms of activity level, $n = 23$, with a mean physical activity level (PAL) of 1.67 ± 0.08 ; *middle*, middle 50% of

our participants, $n = 44$, PAL of 1.55 ± 0.04 ; *lower*, the least active 25% of our participants, $n = 23$, PAL of 1.45 ± 0.05 .

Weight-reduction program

The weight reduction program in this study combined diet counseling with an exercise program. The diet counseling consisted of weekly lectures (90 min per lecture, 12 times in 14 weeks) and individual counseling by skilled dietitians. All subjects were instructed to consume a well-balanced 1200kcal diet per day while also keeping a daily food diary in which they recorded in detail every food they ate during the 14-week intervention period. Skilled dietitians checked the subjects' energy intake (1200kcal) and nutritional balance at every diet counseling class.

The exercise program consisted of some lectures on exercise (60 min per lecture, 3 times in 14 weeks) and supervised exercise sessions (90 min per session, 24 times in 14 weeks). To increase subjects' adherence to the weight reduction program during the intervention period, subjects were assigned to one of two groups taking personal lifestyles into account (occupations, daily schedules, etc.). Participants in group A ($n = 56$) took part in the supervised exercise sessions in addition to the lectures on exercise, while group B participants ($n = 34$) only attended the exercise lectures. However, all subjects could perform free exercise in the fitness gym where the weight reduction program was held so that they were not restricted from performing daily exercise on their own during the intervention period. Our intention was to not control the subjects' daily physical activity (either regular exercise or spontaneous physical activity).

The exercise lectures consisted of the instructions on the basics of exercise, such as the proper way of walking or how to prevent injury during exercise, and all subjects were instructed on how to increase their physical activity during the intervention period. The exercise sessions mainly consisted of aerobic exercise, such as aerobic dance, supervised by physical fitness trainers. Before and after the main exercise session (60 min per session), subjects performed stretching exercises as a warm up or cool down (15 min each).

Statistical analysis

Values are expressed as the mean \pm standard deviation and as mean \pm standard errors. Student paired *t*-tests were performed to test the significance of changes in variables measured before (week 0), during (week 10) or after (week 15) the intervention program. Differences among the three groups for AEE during the intervention were examined using one-way analysis of covariance (ANCOVA) with the baseline BMI representing the covariates, and Tukey–Kramer's post-hoc test applied when the difference was significant ($p < 0.05$) according to the ANCOVA results. The relationship between two

measurement variables was assessed by Pearson's product moment correlation, with values <0.05 regarded as significant. The data were analyzed with the Statistical Analysis System (SAS), version 9.01 (SAS Institute Inc., Cary, NC, USA).

Results

Tables 1 and 2 show subjects' measurement variables from before (week 0), during (week 10) or after (week 15) the intervention program along with changes (*d*) in the measurement variables. Significant reductions were observed in body weight, BMI, waist circumference, IFA, SBP, DBP, TC, TG, LDL-C, FPG, total energy intake, carbohydrate intake, fat intake, protein intake, and TEE. Significant increases were observed in TEE/body-weight and AEE/body-weight.

Table 3 shows correlation coefficients and partial correlation coefficients between weight reduction during the intervention and other measurement variables. Significant correlations were observed between the reduction in body weight and 0 wk body weight, 0 wk BMI, 10 wk TEE, 10 wk AEE, delta (subtraction of 10 wk from 0 wk values) total energy

Table 3 Correlation analysis of body weight reduction with predictors

	Correlation coefficient vs. weight reduction	<i>p</i>	Partial correlation ^a coefficient vs. weight reduction	<i>p</i>
Baseline: week 0				
Body weight	0.45	<0.0001		
BMI	0.43	<0.0001		
During intervention: week 10				
Total energy intake	-0.01	0.96	0.06	0.56
Carbohydrates intake	-0.03	0.78	0.02	0.86
Fat intake	-0.02	0.85	0.02	0.87
Protein intake	-0.02	0.88	0.03	0.83
Total energy expenditure	0.43	<0.001	0.26	0.01
Activity energy expenditure	0.42	<0.001	0.39	<0.001
Delta: subtraction of "week 10" from "week 0"				
Total energy intake	0.27	0.01	0.20	0.06
Carbohydrates intake	0.25	0.02	0.18	0.09
Fat intake	0.17	0.08	0.12	0.27
Protein intake	0.06	0.58	0.03	0.75
Total energy expenditure	-0.18	0.09	-0.11	0.30
Activity energy expenditure	0.02	0.87	0.09	0.42

BMI, body mass index; week 0, before weight reduction program; week 10, at the beginning of the 10th week of the weight reduction program.

^a Partial correlation coefficients adjusted for BMI.

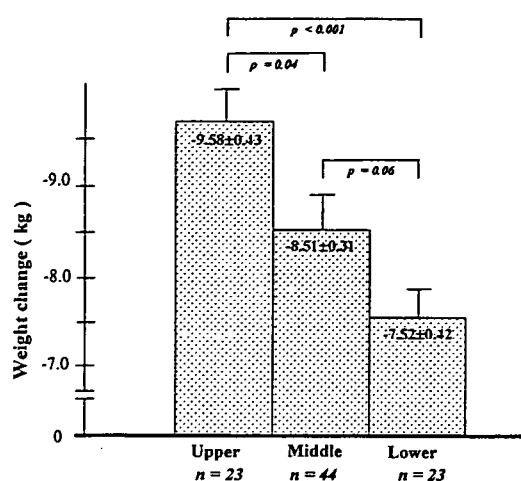


Figure 1 Comparison of weight changes across three categories of activity energy expenditure. Values are presented as the mean \pm standard error. Values are adjusted for baseline BMI. *Upper*, upper level of activity energy expenditure (top 25%); *middle*, middle level of activity energy expenditure (middle 50%); *lower*, lower level of activity energy expenditure (least active 25%).

intake, and delta carbohydrate intake. The correlations of 10 wk TEE and 10 wk AEE with the body weight reduction were also significant when those were adjusted by the 0 wk BMI.

To investigate the relationship between the weight lost and the AEE during the intervention, subjects were assigned to three categories depending on their AEE during the intervention (*upper*: top 25% of participants, $n=23$; *middle*: middle 50%, $n=44$; *lower*: least active 25%, $n=23$). Fig. 1 presents the results of the one-way ANCOVA. Reductions in body weight were compared among the three AEE categories and adjusted for the 0 wk BMI. The decrease in body weight was significantly greater for the subjects in the *upper* AEE classification than for the subjects in the *middle* or *lower* AEE groups. Fig. 2a compares the total energy intake and the intake of each nutrient (carbohydrates, fat, and protein) during the intervention among the three AEE categories adjusted for the 0 wk BMI. Furthermore, Fig. 2b shows the changes in total energy and nutrient intakes before and during intervention, adjusted for the 0 wk BMI, for each of the three AEE categories. No differences across the three AEE categories were observed in any of the analyses.

Fig. 3 shows the relationship between AEE/body-weight before intervention and AEE/body-weight during intervention. The correlation coefficient was 0.57 ($p < 0.0001$). Fig. 4 is a scatter plot showing the percentage of body weight lost relative to the restriction in total energy intake. There

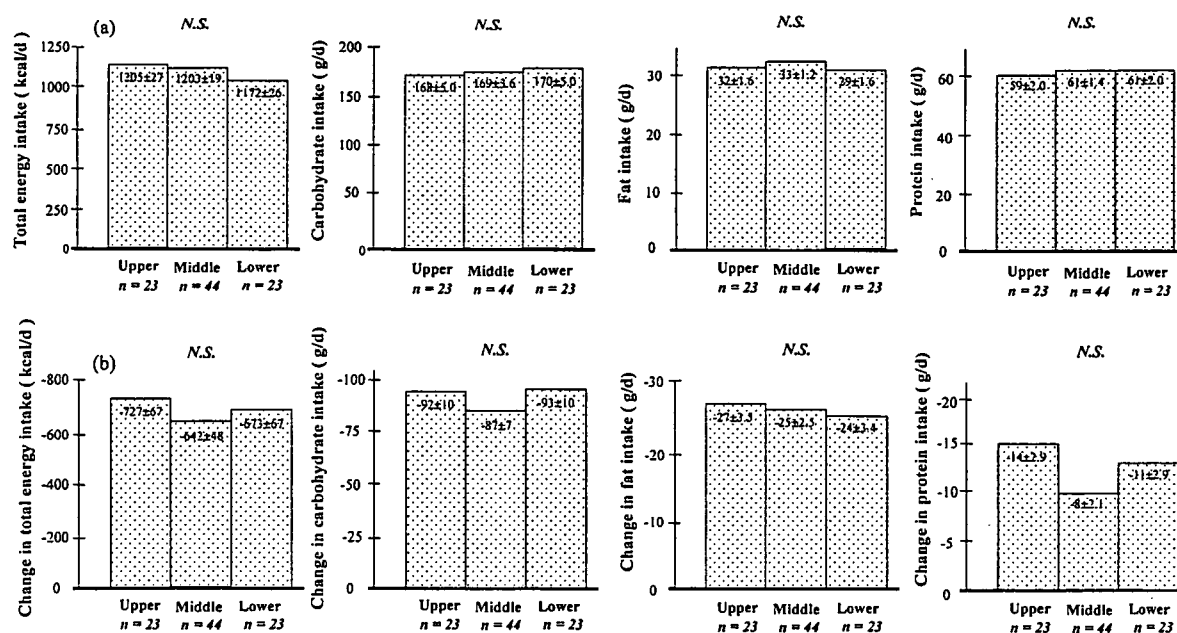


Figure 2 Comparison of the total energy, carbohydrate, fat and protein intakes during the program (a), and comparison of any changes in total energy, carbohydrate, fat and protein intakes (b) across the activity energy expenditure categories. Values are presented as the mean \pm standard error. Values are adjusted for the baseline BMI. *Upper*, upper level of activity energy expenditure (top 25%); *middle*, middle level of activity energy expenditure (middle 50%); *lower*, lower level of activity energy expenditure (least active 25%), N.S., not significant.

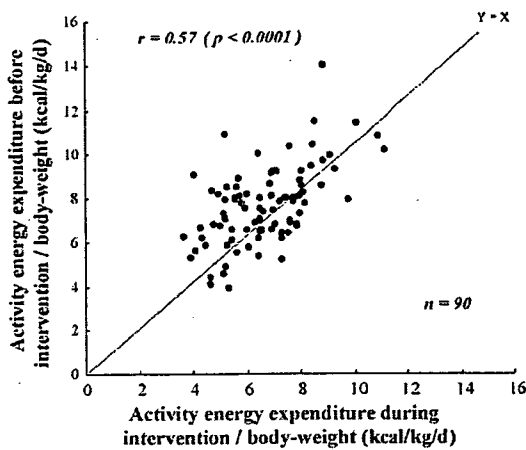


Figure 3 Scatter plot of the activity energy expenditure before and during intervention.

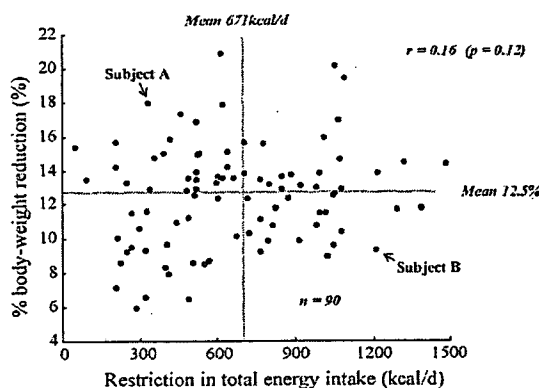


Figure 4 Scatter plot of restriction in total energy intake and % body weight reduction.

was no significant correlation between these two variables.

Discussion

There was no difference in AEE during the intervention between the two exercise groups (group A: 488.4 ± 111.1 kcal/d; group B: 451.5 ± 90.2 kcal/d; data not shown in the table). We considered two possible reasons why there was no difference between the groups: (1) subjects' diaries indicated that some of the group B subjects, which had no supervised exercise sessions, performed free exercise during the intervention period; (2) some studies [18, 19] indicate that voluntary exercise has little impact on total energy expenditure and that spontaneous physical activity has become the major determinant of physical activity energy expenditure. In our study, AEE measured by Lifecorder included not only exercise sessions but also the sub-

jects' daily physical activity; there were subjects whose daily physical activity placed them in the upper level of activity even though they were in group B. In this study, we regarded these two groups as one group.

The reduction in body weight was significantly greater for the subjects whose activity level placed them into the *upper* AEE than for subjects in the *middle* or *lower* AEE (Fig. 1), although there were no dietary differences among the categories (Fig. 2). These results indicate that AEE during the intervention influenced the amount of the body weight lost. Fig. 3 shows the relationship between AEE before and during intervention. A significant correlation ($r=0.57$) was observed between those two variables, indicating that subjects who had a higher level of physical activity during the intervention also had a higher level of physical activity before the intervention. In other words, the subjects who fell into the *upper* AEE category had a more active lifestyle than the subjects within the *lower* AEE category. Although physical activity level during the intervention was related to the reduction in body weight (Fig. 1), this influence was probably due to spontaneous, daily, physical activities, and not due to voluntary exercise. That is, it depended on a subject's daily lifestyle.

Several studies [20–22] have indicated that higher level exercises are necessary to promote body weight reduction. However, we have shown the possibility that ordinary, daily, physical activity, under a strictly controlled diet, can help reduce body weight, and increasing physical activity in one's daily life could play an important role in a weight reduction program.

Levine et al. [23] reported that it is not variable exercise levels, but rather the variance in NEAT (non-exercise activity thermogenesis) that accounts for most of the variability in activity thermogenesis. NEAT is the energy expenditure from all physical activities other than volitional sporting-like exercise [23]. The change in NEAT is predictive of fat gain. Those who, with overfeeding, increase their NEAT the most, gain the least fat, while those who, with overfeeding, do not increase their NEAT, gain the most fat [24]. Our results are consistent with those studies.

On the other hand, it can be difficult to understand why diet restriction alone would not influence body weight reduction. Dunn et al. [25] indicated that dietary changes appeared to be more effective than increased physical activity for weight reduction. It was natural to assume that the dietary restriction in this study (mean decrease of -671 kcal/d) would play an important role in the subjects' loss of body weight. However, we could

find no significant correlation between body weight reduction and diet restriction (i.e. restriction of total energy intake) in this study. Fig. 4 shows the scatter plot of diet restriction relative to body weight reduction. We can see that the reduction of subject A's body weight was greater than the mean value, yet this subject had a less restricted diet than the mean. In contrast, subject B, who lost less weight than the mean, actually had a greater diet restriction than the mean. It is possible that strict diet restriction alone does not always result in a large loss of body weight.

In this study, we investigated the influence of energy intake on body weight reduction in light of diet intake only, but this matter is not so simple. Factors such as digestion, assimilation, metabolism, and evacuation should all be considered in the study of weight loss. Unfortunately, such factors are too complicated to examine them all at this time. Furthermore, Leibel et al. [26] demonstrated that body weight change was not as simple as measuring the energy balance alone. For instance, genetic factors may come into play for people who lose weight easily or with difficulty. Several researchers [27,28] have already reported on the relationship between gene polymorphism and body weight reduction. More approaches to such genetic factors will be needed to solve this issue.

Our study did have two limitations. First, we measured a subject's energy intake during the intervention at just one time (week 10). This one set of intake records was probably not sufficient (3-day WDR) to represent a study period of 14 weeks. However, all subjects still kept food diaries during the 14-week intervention period. They had to record all consumed foods in detail including sugar (g) or oil (ml). Skilled dieticians checked the subjects' energy intakes through the daily diaries at least once every week. We considered most subjects' energy intakes to be approximately 1200 kcal/d during the intervention period, although there were some daily and individual variations.

Second, we assessed subjects' energy expenditures by a uniaxial accelerometry sensor (Lifecorder). Subjects took off the Lifecorder while sleeping or bathing. Since we could not measure EE during these periods, there may have been an underestimation of TEE and AEE. However, the device is becoming an extremely useful method for investigating energy expenditure for epidemiological research [29] because a strong correlation has been observed between Lifecorder readings and calorimetry [16] or the doubly labeled water method [30].

In conclusion, physical activity in daily life in conjunction with a controlled diet can have a positive effect on reducing body weight. This information may be helpful when prescribing a weight management program in obese or overweight women since it is difficult for health advisors to recommend high-intensity exercise programs for them. On the other hand, we did not find any significant correlation between diet restriction and body weight reduction. Our results also suggest that the response to diet restriction tends to differ for each subject. Additional research is needed to increase our understanding of the variability in body weight reduction among individuals so we can develop more effective body weight management programs in the future.

Conflict of interest

The authors of the "The influence of physical activity-induced energy expenditure on the variance in body weight change among individuals during a diet intervention" have no ownership of beneficial interests in such commercial entities, options or warrants to purchase stock or other equity interests, patent-licensing arrangements, or advisory and consulting positions with such commercial entities.

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References

- [1] Rexrode KM, Hennekens CH, Willett WC, Colditz GA, Stampfer MJ, Rich-Edwards JW, et al. A prospective study of body mass index, weight change, and risk of stroke in women. *JAMA* 1997;277:1539–45.
- [2] Kanchaiah S, Evans JC, Levy D, Wilson PW, Benjamin EJ, Larson MG, et al. Obesity and the risk of heart failure. *N Engl J Med* 2002;347:305–13.
- [3] World Health Organization. Obesity: preventing and managing the global epidemic. Geneva, Switzerland; 1997.
- [4] Yoshiike N, Matsumura Y, Zaman MM, Yamaguchi M. Descriptive epidemiology of body mass index in Japanese adults in a representative sample from the National Nutrition Survey 1990–1994. *Int J Obes Relat Metab Disord* 1998;22:684–7.

- [5] Kuzuya T. Prevalence of diabetes mellitus in Japan compiled from literature. *Diabetes Res Clin Pract* 1994;24:S15–21.
- [6] Sakata K, Labarthe DR. Changes in cardiovascular disease risk factors in three Japanese national surveys 1971–1990. *J Epidemiol* 1996;6:93–107.
- [7] JASSO. Guidelines for diagnosis and treatment in obesity and its comorbidities. The 20th Meeting, October; 1999.
- [8] Rippe JM, McInnis KJ, Melanson KJ. Physician involvement in the management of obesity as a primary medical condition. *Obes Res* 2001;9:S302–11.
- [9] Okura T, Tanaka K, Nakanishi T, Lee DJ, Nakata Y, Wee SW, et al. Effects of obesity phenotype on coronary heart disease risk factors in response to weight loss. *Obes Res* 2002;10:757–66.
- [10] Okura T, Nakata Y, Yamabuki K, Tanaka K. Regional body composition changes exhibit opposing effects on coronary heart disease risk factors. *Arterioscler Thromb Vasc Biol* 2004;24:923–9.
- [11] Tanaka K, Okura T, Shigematsu R, Nakata Y, Lee DJ, Wee SW, et al. Target value of intraabdominal fat area for improving coronary heart disease risk factors. *Obes Res* 2004;12:695–703.
- [12] Eilat-Adar S, Eldar M, Goldbourt U. Association of intentional changes in body weight with coronary heart disease event rates in overweight subjects who have an additional coronary risk factor. *Am J Epidemiol* 2005;161:352–8.
- [13] Donnelly JE, Smith BK. Is exercise effective for weight loss with ad libitum diet? Energy balance, compensation, and gender differences. *Exerc Sport Sci Rev* 2005;33:169–74.
- [14] Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972;18:499–502.
- [15] Science and Technology Agency. Standard tables of food composition in Japan. 4th revised ed. Tokyo: Printing Bureau. Ministry of Finance; 1982 [in Japanese].
- [16] Kumahara H, Schutz Y, Ayabe M, Yoshioka M, Yoshitake Y, Shindo M, et al. The use of uniaxial accelerometry for the assessment of physical-activity-related energy expenditure: a validation study against whole-body indirect calorimetry. *Br J Nutr* 2004;91:235–43.
- [17] Borg GA. Perceived exertion: a note on "history" and methods. *Med Sci Sports* 1973;5:90–3.
- [18] Goran MI, Poehlman ET. Endurance training does not enhance total energy expenditure in healthy elderly persons. *Am J Physiol* 1992;263:950–7.
- [19] Thorburn AW, Proietto J. Biological determinants of spontaneous physical activity. *Obes Rev* 2000;1:87–94.
- [20] Jeffery RW, Wing RR, Sherwood NE, Tate DF. Physical activity and weight loss: does prescribing higher physical activity goals improve outcome? *Am J Clin Nutr* 2003;78:684–9.
- [21] Ross R, Janssen I, Dawson J, Kungl AM, Kuk JL, Wong SL, et al. Exercise-induced reduction in obesity and insulin resistance in women: a randomized controlled trial. *Obes Res* 2004;12:789–98.
- [22] Slentz CA, Duscha BD, Johnson JL, Ketchum K, Aiken LB, Samsa GP, et al. Effects of the amount of exercise on body weight, body composition, and measures of central obesity: STRRIDE—a randomized controlled study. *Arch Intern Med* 2004;164:31–9.
- [23] Levine JA, Vander Weg MW, Hill JO, Klesges RC. Non-exercise activity thermogenesis: the crouching tiger hidden dragon of societal weight gain. *Arterioscler Thromb Vasc Biol* 2006;26:729–36.
- [24] Levine JA, Eberhardt NL, Jensen MD. Role of nonexercise activity thermogenesis in resistance to fat gain in humans. *Science* 1999;283:212–4.
- [25] Dunn CL, Hannan PJ, Jeffery RW, Sherwood NE, Pronk NP, Boyle R. The comparative and cumulative effects of a dietary restriction and exercise on weight loss. *Int J Obes* 2006;30:112–21.
- [26] Leibel RL, Rosenbaum M, Hirsch J. Changes in energy expenditure resulting from altered body weight. *N Engl J Med* 1995;332:621–8.
- [27] Aberle J, Evans D, Beil FU, Seedorf U. A polymorphism in the apolipoprotein A5 gene is associated with weight loss after short-term diet. *Clin Genet* 2005;68:152–4.
- [28] Corella D, Qi L, Sorli JV, Godoy D, Portoles O, Coltell O, et al. Obese subjects carrying the 11,482G > A polymorphism at the perilipin locus are resistant to weight loss after dietary energy restriction. *J Clin Endocrinol Metab* 2005;90:5121–6.
- [29] Yoshioka M, Ayabe M, Yahiro T, Higuchi H, Higaki Y, St-Amand J, et al. Long-period accelerometer monitoring shows the role of physical activity in overweight and obesity. *Int J Obes* 2005;29:502–8.
- [30] Rafamantanantsoa HH, Ebine N, Yoshioka M, Higuchi H, Yoshitake Y, Tanaka H, et al. Validation of three alternative methods to measure total energy expenditure against the doubly labeled water method for older Japanese men. *J Nutr Sci Vitaminol (Tokyo)* 2002;48:517–23.

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軽度要介護者の血中ビタミン D レベルの分布状況と ビタミン D・乳酸カルシウム製剤補充による介護予防効果

—生活機能・身体機能と血中ビタミン D レベルとの関連より—

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はじめに

介護保険制度が施行され 5 年がたち、要介護認定者が約 2 倍に増加している。全体の約 5 割を占める要支援・要介護 1 の軽度要介護者は、サービスが開始された 2 年後には半数以上が重度化しており、サービスが利用者の状態改善につながっていないといわれている。平成 13 年国民生活基礎調査によると介護が必要となった主な疾患は骨折・転倒、関節疾患（リウマチ等）で 27～28% を占めている。在宅の軽度要介護者の多くは、関節疾患などがあるために下肢機能の低下が目立ち、要介護の主要原因としてあげられている転倒・骨折の危険性が高い集団であり、これらを予防することが介護度悪化を予防できると推測される。

ビタミン D の欠乏は、骨粗鬆症の危険因子であり、高齢者のふらつきや転倒と関連があり^{1,2)}、筋力の低下³⁾や高齢者の下肢機能低下⁴⁾をもたらすといわれており、筋力低下は高齢者の転倒の主要な危険因子である⁵⁾。ビタミン D の補充は、骨密度を増加させる作用の他に、転倒の予防やふらつき・つまずきを改善するという報告がある^{1,2)}。一方、効果がみられないという報告もあり⁶⁾、ビタミン D の評価は一定ではない。

われわれはこれまで地域在住の軽度要介護者と特定高齢者を対象にビタミン D 濃度と生活機能・身体機能との関連を研究してきたので報告す

る。

1 目 的

本研究では、①要支援・要介護 1 の高齢者・特定高齢者の血清ビタミン D 濃度と身体機能との関連を横断調査により明らかにする。また、②ビタミン D・乳酸カルシウム製剤の補充による身体機能への効果を縦断的に検討することを目的とした。

2 方 法

1) 対 象

平成 17・18 年の 6～9 月に介護予防教室に参加した茨城県 Y 町（北緯 36 度）の地域在住の要支援・要介護 1 の 65 歳以上の高齢者（軽度者）、特定高齢者レベルの者（基本チェックリストの「運動」の項目が 3 個以上該当）61 名。

2) 調査方法

質問紙による面接聞き取り調査、体力測定、採血を行った。

①質問項目：属性、生活機能、ADL (Barthel index)、転倒回数、つまずき・ふらつきの有無、外出回数、椅子やベッドからの起き上がり・立ち上がりなどを調査した。

②体力測定：歩行能力として Timed Up & Go (TUG)・5m 歩行、柔軟性として長座体前屈、バランス能力としてファンクショナルリーチ・開眼

Key words : 軽度要介護者, ビタミン D, 乳酸カルシウム製剤

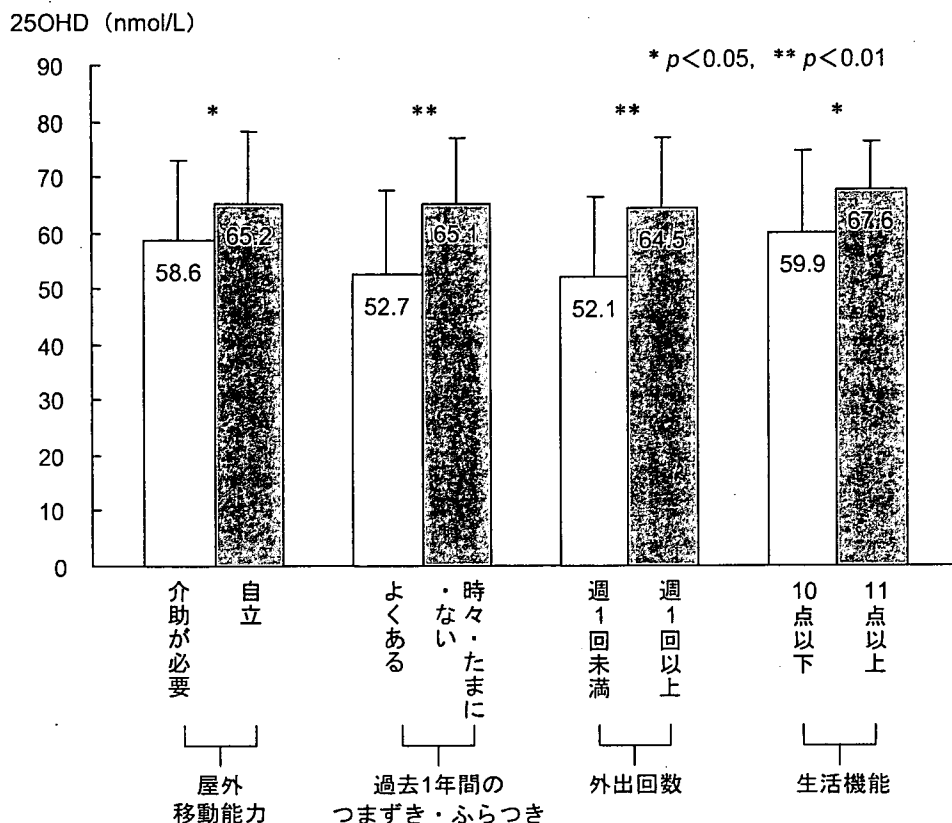


図 1 血清ビタミン D と関連のある項目

片足立ち、筋力として握力・足関節背屈力を測定した。2回測定し平均値を用いた。測定が1回の者はその値を用いた。開眼片足立ち・握力・足関節背屈力は左右の平均値を用いた。

③血液データ: 血清アルブミン, クレアチニン, カルシウム, intact PTH (iPTH) (ECLIA 法), ビタミン D (25OHD) (RIA 法) を測定した。

上記の調査は初回と3ヵ月目に実施した。

④介護予防教室: 運動と栄養指導を合わせた包括的なプログラムよりなり、週1回約90分12回開催した。参加者のうち38名は介護予防教室へ参加し(運動群), 残りの23名は介護予防について説明を受けただけである(コントロール群)。

⑤ビタミン D・乳酸カルシウム製剤の補充効果: 運動群の希望者 (n=17) にアルファカルシドール 1μg/日および乳酸カルシウム 4g/日を3ヵ月間投与した。服用1ヵ月目に採血を実施し副作用チェックを行った。

3) 解析方法

2群間の比較には、連続変数の場合は t 検定を、

カテゴリ変数の場合は χ^2 検定または Fisher の直接法により比較検討した。また、多重ロジスティック回帰分析により「過去1年間のつまずき・ふらつき」に影響する要因を検討した。ビタミン D 製剤服用有無の影響は繰り返しのある2元配置分散分析により解析した。また、開始前後の連続変数の比較は paired t-test を実施した。統計解析には SPSS 12.0 J for Windows を使い、 $p < 0.05$ を有意差ありとした。

本研究は、筑波大学人間総合科学研究科の倫理委員会の承認を得、参加者には文章と口頭による説明を行い同意を得てから実施した。

3 結 果

1) 対象者の背景

参加者 61 名 (運動群とコントロール群) の開始時の特性は、平均年齢: 77.0±5.6 歳 (65~90 歳), 男性: 18 名 (29.5%), Barthel index 平均得点: 89.7±10.3, 過去1年間に転倒経験有: 32 名 (52.5%), つまずき・ふらつき経験有: 45 名

表 1 ビタミン D・乳酸カルシウム製剤服用有無による身体機能への効果—運動群において—

	ビタミン D 非服用群 (n=16)	ビタミン D 服用群 (n=16)	交互作用
25OHD (nmol/L)	59.9±11.9	65.1±14.8	
pre TUG (sec)	17.1±8.2 ^a	23.4±8.0	
post TUG (sec)	14.4±7.9*	18.6±5.1**	
pre 5m 通常歩行 (sec)	8.2±3.3	8.5±2.8	
post 5m 通常歩行 (sec)	7.2±4.7	8.3±2.4	
pre 長座体前屈 (cm)	7.6±8.1	3.0±6.4	
post 長座体前屈 (cm)	6.6±11	3.7±6.7	
pre ファンクショナルリーチ (cm)	21.6±5.5	20.6±5.0	
post ファンクショナルリーチ (cm)	24.2±5	22.1±6.5	
pre 開眼片足立ち ^b (sec)	9.1±11.7	4.0±2.6	
post 開眼片足立ち (sec)	11.3±13.1	5.8±4.6*	
pre 握力 ^b (kg)	17.7±8.7	19.6±7.4	
post 握力 (kg)	19.7±7.5	21.2±6.3	
pre 足関節背屈力 ^b (kg)	8.7±2.1	10.7±3.6	#
post 足関節背屈力 (kg)	9.4±2.1	13.4±3.7**	

Values are Means±SD, TUG : Timed Up & Go, * $p < 0.05$ vs pre, ** $p < 0.01$ vs pre,

a : $p < 0.05$ vs ビタミン D 服用群

b : 開眼片足立ち・握力・足関節背屈力は左右の平均値を用いた。

: $p < 0.05$

(73.8%), 血清 25OHD 濃度 (±SD) : 62.0±14.0 nmol/L (27.5~87.5), 血清 25OHD<50nmol/L の割合:18.0%, 血清 iPTH (±SD) : 48.7±22.5pg/mL (17.0~118.0) であった。年齢・性で調整後も、血清 25OHD は iPTH と有意な負の相関 ($r = -0.38$, $p < 0.01$) を示していた。

2) 血清 25OHD と生活機能・身体機能との関連

一人で歩ける者は支えが必要な者に比し、週 1 回以上外出する者は未満の者に比し、生活活動能力指標総得点が 11 点以上の者は 10 点以下の者に比し、25OHD 濃度は有意に高い値を示し、過去につまづき・ふらつきがよくあると回答した者はその他の者に比し、25OHD 濃度が有意に低い値を示した (図 1)。また、「つまづき・ふらつきがよくある」に影響する因子を年齢・性で調整した多重ロジスティック回帰分析を実施した結果、25OHD 濃度が独立した影響因子であった (OR : 0.92, 95%信頼区間 0.87~0.97)。

3) ビタミン D・乳酸カルシウム製剤服用有無と身体機能との関連

運動群 ($n = 38$) の希望者 ($n = 17$) にビタミン D としてアルファカルシドール $1 \mu\text{g}/\text{日}$ と乳酸カルシウム $4\text{g}/\text{日}$ を 3 ヶ月間投与し (服用群), 希望しなかった者は非服用群 ($n = 21$) とした。開始時と 3 ヶ月目の身体測定データがある者を解析対象とし (服用群 16 名, 非服用群 16 名), 身体機能との関連を検討した (表 1)。服用群と非服用群間で開始時の 25OHD 濃度, 25OHD<50 nmol/L の割合, TUG 以外の体力測定値に有意差はみられなかった。開始時の TUG は服用群のほうが非服用群より有意に劣っていたが, 3 ヶ月目には有意差はみられなかった。1 ヶ月目の採血で副作用があった者はいなかった。ビタミン D・乳酸カルシウム投与の効果を繰り返しの 2 元配置分散分析を行った結果, 足関節背屈力にビタミン D・乳酸カルシウム投与有無と時間との間に

交互作用がみられたが、その他の項目ではみられなかった。そこで、各群において、開始時と 3 ヶ月目の身体機能を比較検討した。服用群では、TUG・開眼片足立ち・足関節背屈力が有意に改善し、非服用群においても TUG が有意に改善していた。さらに、開始時の 25OHD 濃度がどのように影響しているか各群で検討を試みたところ、50nmol/L 以上の者は有意に改善していたが、50 nmol/L 未満の者は数が少なく統計的な解析は困難であったが、身体機能の改善はみられない傾向であった。

4 考 察

ビタミン D の欠乏は、転倒・骨折、下肢機能の低下や筋力の低下と関連があると報告されている。本研究の目的①として、起き上がり・立ち上がり・歩行能力の低下が特徴とされている地域在住の軽度要介護者と特定高齢者を対象に血清ビタミン D 濃度と ADL、身体機能との関連について横断的に検討した。屋外での移動能力として「支えが必要・一人で移動ができない者」、「過去 1 年間のつまずき・ふらつきがよくある者」、「外出回数が週 1 回未満の者」、「生活機能得点が 10 点以下の者」は、その他の者に比し、25OHD 濃度は有意に低い値であり、また、25OHD 濃度が 1nmol/L 上昇すると「つまずき・ふらつき」が 8% 低下することから、25OHD 濃度は生活機能・歩行能力・バランス能力と関連があることが示唆された。

本研究の対象者の 52.5% は過去 1 年間に 1 回以上の転倒経験があり、日本の地域在住高齢者の 1 年間の転倒発生率である約 10~20%⁷⁾と比較すると、本研究対象者のほうが転倒歴のある者の割合が高かった。さらに、約 74% が過去 1 年間につまずき・ふらつきの経験があり、将来、転倒・骨折へつながる可能性が非常に高い集団であることが推測されることから、軽度要介護者や特定高齢者の 25OHD 濃度を測定することは介護予防にとって重要と思われた。

ビタミン D 補充の効果に関しては一致した見解はない^{4,6)}。さらに、日本の介護保険対象者への

ビタミン D 補充による介護予防効果に関する研究はほとんどみあたらない。本研究では目的②のビタミン D の補充効果について、介護予防教室に参加した運動群の希望者にビタミン D としてアルファカルシドール 1 μ g/日、乳酸カルシウム 4g/日を 3 ヶ月間投与し（服用群）、身体機能への効果を服用しなかった群（非服用群）と比較検討した。繰り返しのある 2 元配置分散分析により分析した結果、3 ヶ月目の足関節背屈力は開始時に比し、ビタミン D・乳酸カルシウム製剤補充により有意に改善し、ビタミン D・乳酸カルシウムの補充は、脚筋力を改善し、将来の転倒を予防できる可能性が示唆された。しかし、その他の身体機能に関しては、ビタミン D・乳酸カルシウム製剤服用有無で身体機能に交互作用はみられなかった。そこで、各群で教室前後の身体機能を比較した結果、服用群では TUG・開眼片足立ち・足関節背屈力が有意に改善した。さらに、非服用群でも TUG が有意に改善しており、その理由の一つとして、開始時の 25OHD 濃度が 50nmol/L 未満だと 3 年後には体力が低下しているという報告があるように⁸⁾、開始時のビタミン D 濃度がその後の体力に影響している可能性が考えられる。また、先のわれわれの横断研究により⁹⁾、歩行能力やふらつきとの関連からビタミン D 不足の閾値を 25OHD < 50nmol/L と設定した場合、各群とも開始時の 25OHD が 50nmol/L 未満の者は教室終了時にも身体機能は改善しない傾向を示していた。開始時の 25OHD 濃度を少なくとも 50nmol/L 以上維持し、軽い運動を継続することで下肢筋力、歩行能力を維持改善できる可能性が推測されるが、本研究は対象者数が少ないことから、今後さらに対象者を増やし検討する必要がある。

結 論

地域在住の軽度要介護者・特定高齢者を対象とした場合、低い 25OHD 濃度は、「閉じこもり」高齢者、生活機能が低下している者、移動能力が劣っている者、バランス能力が低下している者と関連があった。また、運動の提供と同時にビタミン D・乳酸カルシウム製剤の補充は歩行能力・バ

ランス能力・下肢筋力を改善することが示唆された。

文 献

- 1) Pfeifer M, Begerow B, Minne HW, Abrams C, Nachtigall D, Hansen C. Effects of a short-term vitamin D and calcium supplementation on body sway and secondary hyperparathyroidism in elderly women. *J Bone Miner Res* 2000;15:1113-8.
- 2) Bischoff HA, Stahelin HB, Dick W, Akos R, Knecht M, Salis C, et al. Effects of vitamin D and calcium supplementation on falls: a randomized controlled trial. *J Bone Miner Res* 2003;18:343-51.
- 3) Bischoff HA, Stahelin HB, Urscheler N, Ehrensam R, Vonthein R, Perrig-Chiello P, et al. Muscle strength in the elderly: its relation to vitamin D metabolites. *Arch Phys Med Rehabil* 1999;80:54-8.
- 4) Gerdhem P, Ringsberg KA, Obrant KJ, Akesson K. Association between 25-hydroxy vitamin D levels, physical activity, muscle strength and fracture in the prospective population-based OPRA Study of Elderly Women. *Osteoporos Int* 2005;16:1425-31.
- 5) Moreland JD, Richardson JA, Goldsmith CH, Clase CM. Muscle weakness and falls in older adults: a systematic review and meta-analysis. *J Am Geriatr Soc* 2004;52:1121-9.
- 6) Porthouse J, Cockayne S, King C, Saxon L, Steele E, Aspray T, et al. Randomised controlled trial of calcium and supplementation with cholecalciferol (vitamin D₃) for prevention of fractures in primary care. *BMJ* 2005;330:1003-9.
- 7) 新野直明. 総括研究報告書 平成 11 年度厚生労働省長寿科学総合研究「地域の高齢者における転倒・骨折の発生と予防に関する疫学的研究」報告書 (主任研究者: 新野直明). 1999.
- 8) Wicherts IS, van Schoor NM, Boeke AJ, Visser M, Deeg DJ, Smit J, et al. Vitamin D status predicts physical performance and its decline in older persons. *J Clin Endocrinol Metab* 2007;92:2058-65.
- 9) 奥野純子, 戸村成男, 柳久子. 地域在住虚弱高齢者のビタミン D 濃度の分布状況とビタミン D 濃度と生活機能・身体機能との関連. *日老医誌* 2007; 44:634-40.