

HEALTH AND NUTRITION NEWS

No. 19

March 15, 2007



Table of Contents

PREFATORY NOTE

Current issues on nutrition in the clinical field; e.g. lifestyle-related diseases.....**Tsuyoshi Watanabe**

CURRENT RESEARCH PROJECTS

Project for Energy Metabolism.....**Kazuko Ishikawa-Takata**

THOUGHTS ON HEALTH AND NUTRITION RESEARCH

Traveling the universe of gene.....**Kouichi Yamada**

RESEARCH FINDINGS

AMP-activated protein kinase regulates PEPCK gene expression by direct phosphorylation of AREBP
.....**Erina Inoue, Jun Yamauchi**

Young Japanese adults with high BMI tend to underreport energy intake – from the dietary records of
the general population aged 30's-60's.....**Hitomi Okubo, Satoshi Sasaki**

Effects of age on ventilatory threshold and peak oxygen uptake normalized for regional skeletal
muscle mass: a cross-sectional study on Japanese men and women aged 20-80 years
..... **Motohiko Miyachi**

Prefatory Note

Current issues on nutrition in the clinical field; e.g. lifestyle-related diseases

Tsuyoshi Watanabe

Professor, Department of Internal Medicine 3 (renal disease, hypertension, diabetes, internal secretion, metabolic disorder)
School of Medicine, Fukushima Medical University



Now, lifestyle-related diseases are the major social problems in Japan, which include conventionally called as “adult diseases (e.g, diabetes, hypertension, hyperlipidemia etc.)” and recently well-advocated metabolic syndrome. The above diseases would accompany serious complications such as atherosclerosis, diabetic nephropathy, diabetic retinopathy, neurosis, nephrosclerosis (caused by hypertension), end-stage renal disease, blindness and intolerable neurologic symptom, leading to the lowered QOL among the population. In particular, atherosclerosis is the base of cardiovascular event, which is one of the top mortality causes with the overwhelmingly highest morbidity rate in the world.

Besides, with low economic growth and a fear of bankruptcy of the public finances for medical insurance due to the aging society, the proportion of the expense for lifestyle-related diseases and their complications in the national medical expenditures has been increasing, and now exceed other types of health problems. In this sense, the government decided to set lifestyle-related diseases control as the most prioritized national health strategy in the 21st century, where “adult disease” was renamed “lifestyle-related diseases”.

Basically, susceptibility to lifestyle-related diseases is genetically determined. Studies on the genetic background have elucidated that diabetes, hypertension and hyperlipidemia are caused by single-gene abnormalities, though they are exceptional. In fact, most of lifestyle-related diseases are polygenic, and the responsible genes were not selected for elimination in the process of human evolution (history of hunger), but rather worked favorably for preservation of species (so-called “thrifty genes”). Consequently, the genetic

background of many Japanese people would exceed the threshold level for the occurrence of lifestyle-related diseases. In particular, Japanese diet has been composed mainly of plants since the beginning of human history. It is therefore likely that the genetic background with low digestive and absorptive capacity was selected, in which one would develop lifestyle-related diseases by high energy and fat intake. It is also pointed out that Japanese people have relatively longer small intestine than meat-eating western people. We should therefore bear in mind that Japanese is the ethnic group who fatefully carry lots of thrifty genes.

Needless to say, a dramatic increase of lifestyle-related diseases is not caused by the change in genetic factors. After the World War II, Japan experienced high economic growth and became the world's second-largest economy. As a result, the general people could enjoy rich food supply which was once accessible by the royalty and aristocracy only. In particular, westernization of lifestyle shifted food preference from staple food, vegetable and fish to meat, oil/lipid and milk, leading to establishment of the westernized gourmet culture. This trend can be observed also in the annual National Nutrition Survey (renamed to National Health and Nutrition Survey in 2003) undertaken by the Ministry of Health, Labour and Welfare, which shows that the nutrient intake has been dramatically changed with decreased carbohydrate, increased fat and slightly increased protein intake. It is said that high fat diet, especially excess intake of animal fat that contains high saturated fatty acid, could induce visceral adiposity with enhanced insulin resistance, which eventually cause lifestyle-related diseases. No significant change has been observed in the average energy intake per capita during the

postwar period. Yet, many of general people still express “living” as “eating”, which implicates that the past hunger experience would unconsciously make us to seek for gourmet diet (high fat diet). Hence, the average Body Mass Index (BMI) of Japanese people has been increasing, except for young women. On the other hand, there has been a decreasing trend in the average BMI of young women, possibly due to dieting with the desire of being slim, and some of them even develop anorexia nervosa. In addition, the decreased physical activity due to development of transportation infrastructure and household electric appliances would increase the risk of developing lifestyle-related diseases. Physical activity used to be a measure (not an objective) to establish living environment and to secure necessary food, while the current low physical activity would reflect affluent and affluent livings where one’s frustration can be released by sports, game and even by fighting only. As a result, body fat is accumulated (obesity), especially visceral fat, by excess energy intake which does not match physical activity level. It is therefore obvious that the changes in our lifestyle, especially food culture, have contributed to substantial increase of lifestyle-related diseases. Now, how the problems on diet can be solved in the clinical field? Historically, human beings continued to pursue the missions to secure living environment that protect us against bacteria, animal, severe climate, as well as food that protect us against hunger and undernutrition, seeking for good health. However, the progress of scientific technology, especially after the industrial revolution, has dramatically improved food production and distribution system, leading to the change of whole situation. In the 20th century, the scientific technology was mostly utilized for various wars and colonial rule began. As a negative asset of the 20th century, many developing countries still suffer from inadequate living environment and hunger in the 21st century too. On the other hand, the developed countries also face with the global environmental problems (e.g. environmental prolusion, global warming etc.), which can not be simply solved by the advancement of scientific technology. Well, what did the easy access to nutrient food bring to our health, then? Thanks to improvement in availability

of nutrient food and advancement in medical science and health system, infectious diseases and undernutrition were controlled and average life expectancy was prolonged. Consequently, Japan became the aging society with the world longest average life expectancy after the WWII. Apart from the financial problem on medical insurance, at the individual level, it is good to secure one’s longevity. On the other hand, the negative aspect came up at the individual level as well, that is, lifestyle-related diseases. Most of lifestyle-related diseases were classified as “adult diseases” in the past, as the morbidity of these diseases increased with age. Thus, the aging society is one of the factors responsible to the increase of lifestyle-related diseases. In other words, we can regard lifestyle-related diseases as a “payback” of social progress. However, we should not sort out this problem by simply stepping back in our history, which is practically impossible. Now, there are many issues for the medical professionals to work out including; 1) treatment of lifestyle-related diseases and their complications, 2) interventions for improvement of lifestyle behaviors, which does not make the patients uncomfortable, 3) identification of risk group by elucidation of the genetic background, and 4) development of safe and effective therapeutic drugs, which could also contribute to improvement of lifestyle behaviors.

I think that the clinical nutrition should work on the solutions for lifestyle-related diseases under the current comfortable aging society. As a clinician in charge of lifestyle-related diseases, I would like to work out these issues multilaterally, in cooperation with the professionals in other fields.

** The original Japanese version was translated by Project for International Research and Development, NIHN*



Current Research Projects

Project for Energy Metabolism

Kazuko Ishikawa-Takata

Health Promotion and Exercise Program

Currently, I belong to Project for Energy Metabolism in Health Promotion and Exercise Program. In this project, I have been working on the researches on energy metabolism and physical activity. The main research objectives are as below;

- (1) To measure energy expenditure and physical activity level of various activities, and to identify the influencing factors.
- (2) To develop a simple methodology to assess energy expenditure and physical activity level.
- (3) To investigate the association with developing disease and maintaining the capacity to live independently among the elderly, in terms of both physical activity and nutrition.

For the research (1), we measure energy expenditure in daily life, using doubly-labeled water (DLW) method, in which a stable isotope of hydrogen and oxygen is used. This isotope is included in drinking water too, though the amount is very little. In DLW method, the subjects are asked to take DLW, which contains more stable isotopes than drinking water, and also to collect their urine once a day for 1-2 weeks. Using the urine samples, the amount of CO₂ produced from our bodies can be measured, and then energy expenditure can be calculated. The main advantages of this method include; 1) No restriction with the activities for the subjects, 2) The required tasks are not too much (only urine collection and record of collection time), 3) precision of the data is secured. Therefore, the measurement can be made regardless of one's age, from young infants to the elderly. In this way, energy expenditure of various types of subjects, even the athletes, can be measured using DLW method. In addition, we investigate the factors by which the daily energy expenditure could change; e.g. type of activity, body composition, diet and season.

The research (2) is carried out as a joint study with other projects of our institute, university staffs and private companies. Whilst it has been well acknowledged that DLW method is precise, its cost is very high and it is also difficult to measure many subjects. Besides, we can obtain only the average data of energy expenditure for the 1-2 weeks. Therefore, a simple and precise method to measure

physical activity level per day and for each activity is required. Now, there are the equipments by which we can see our own energy expenditure (e.g. pedometer). Yet, its accuracy is still questioned and it is necessary to develop more precise equipments. Furthermore, a simple and precise method to measure energy intake is also urgently required. DLW method could provide the basic data to investigate the above aspects as well.

Utilizing the data obtained in the above (1) and (2), the research (3) examines the necessary physical activity level and energy expenditure for prevention of disease development and of decrease in the capacity to live independently, with respect to both the contents of energy expenditure and nutrient intake. In addition, we have a collaborative study with National Center for Geriatrics and Gerontology, in which we investigate the factors associated with the decreased capacity to live independently, so as to propose adequate physical activity and diet.

Since my researches are basically consisted of human studies, I appreciate the understanding and cooperation of the study subjects as well as the cooperation by many staff in other projects/institutes.

Energy metabolism is based on energy intake, utilization and expenditure, which is associated with all of nutrition, physiology and exercise. I would like to pursue the researches that would contribute to maintaining one's health and the capacity to live independently, in terms of both diet and exercise.



Equipment for pre-treatment of samples in DLW method



Stable isotope ratio mass spectrometer

Thoughts on Health and Nutrition Research

Traveling the universe of gene

Kouichi Yamada
Nutritional Education Program

When I was looking for the information on lifestyle-related diseases on the internet, I noted a website on dieting which says "Let's check your obesity gene!". That is, if one sends the fingernails or cotton swab that wiped inner mouth to the inspection institute by post, and one's polymorphism in a gene can be analyzed. Three obesity genes are examined by this method; beta 2 adrenergic receptor (β 2AR), beta 3 adrenergic receptor (β 3AR) and uncoupling protein 1 (UCP1). In this website, it is also written as follows; if one has gene mutation (exactly, alteration) in UCP1, s/he is susceptible to subcutaneous obesity (pear-shaped). Twenty-five percent of Japanese have the mutation in UCP1, and their energy metabolism is 100kcal lower than that of people without the mutation. Women are more likely to develop this type of obesity, with having a lot of subcutaneous fat in hip and thigh. I was actually surprised to see the advice written in the website, which says that "you should take staple food regularly and less fatty foods, and also take in the order of vegetable, staple food and meat/fish". Indeed, we are still studying to verify the accuracy of information as well as the evidences.

Locations of single-nucleotide polymorphism (SNP) to be analyzed are Arg16Gly for β 2AR, Trp64Arg for β 3AR and Met229Leu for UCP1, according to the above website. As for UCP1, -3826A>G has been well examined since before, though most of which reported no association with weight gain. Practically, it should be questionable UCP1 gene expression can be attenuated by SNP located 4kb upstream. Nobody has measured the promoter activity of this region. Mori et al. reported the Met229-->Leu as well as -112A-->C minor allele were frequently observed in the type II diabetes patients, and examined influence of -112 SNP on UCP1 promoter activity using the reporter assay (*Diabetologia* 2001, 44, 373-6). They also reported the linkage disequilibrium between -112 and Met229, which shows that -112 may be more likely than Met229 to be the causative SNP. In this way, the

cause-effect relationships between SNPs in UCP1 and predisposition to obesity have not been fully elucidated yet.

The above mentioned "big-three" genes and a peroxisome proliferator-activated receptor gamma ($PPAR\gamma$ 2) are known as "four kings" of obesity genes. Yet, in practice, hereditary factors of obesity cannot be simply explained by these four genes only, as more than 50 genes are known to be related to obesity. Besides, there is a new gene that has been recently drawn attention, that is, RAGE (advanced glycosylation end product-specific receptor gene). Well, I could not understand what it meant, when I heard of it for the first time. Later, searching in the databases of Entrez-Nucleotide and Entrez-Gene, I learned that RAGE was the gene located in human chromosome 6p21.3 and I could also view its exon-intron structure. I am amazed to see the great gap between the current advanced technology and the past one where it took several days for sequencing even dozens or hundreds base by Maxam-Gilbert method or dideoxy method.

RAGE is the gene that encodes 404 or 302 amino acid residue proteins. It is not so large gene, but is divided into 11 exons. This number is rather big for its size. For example, β 2AR and β 3AR have only 1 exon and 2 exons, respectively. Besides, RAGE is surrounded by many genes like a cluster housing, whereas the genes around β 2AR are scattered with dozens kb of distance to the nearest gene. Likewise, β 3AR also has only 2 genes within 100kb distance. Um... our creator is really full of whims, isn't it?

When I look at a large amount of base sequences in the SNP analyses, I feel that they look like mantra or secret codes. Exons account for only 2% of human genome (300 millions bp), and the protein-coding region is only 1.1%. Intron accounts for 24% and the remainder is meaningless intergenic region. So, exon looks like a green oasis in desert or even like a water planet floating in the universe. In order to search unknown metabolic code, I would continue to travel the universe of gene hereafter too

AMP-activated protein kinase regulates PEPCK gene expression by direct phosphorylation of AREBP

Erina Inoue, Jun Yamauchi (Nutritional Epidemiology Program)

ATP-activated protein kinase (AMPK) acts as an intracellular sensor for maintaining the energy balance, and is activated by hypoglycemia and exercise in skeletal muscle. The activated AMPK maintains the energy balance by direct phosphorylation of major metabolic enzymes for glucose and lipid and by changing these enzyme activities. In addition, it has been recently proposed that the activated AMPK regulates gene expression, and thus AMPK may directly affect activation of transcriptional factors.

Blood glucose is important energy source in human bodies, which is maintained at a certain level by keeping the balance between hepatic gluconeogenesis and uptake of glucose by peripheral tissue. Excessive hepatic glucose output through abnormally high levels of glycogenolysis and gluconeogenesis as well as poor uptake glucose uptake are characteristics of diabetes. Gluconeogenesis is regulated by the key enzymes such as phosphoenolpyruvate carboxykinase (PEPCK) and glucose-6-phosphatase. PEPCK catalyzes the irreversible conversion during the initial reaction in hepatic gluconeogenesis. Thus, PEPCK gene expression is especially important, which is mediated by several circulating hormones such as glucagons and glucocorticoids, whereas it is suppressed by insulin. In the patients with insulin resistant diabetes, PEPCK gene expression cannot be suppressed by insulin, leading to the vicious cycle for elevated blood glucose. Therefore, another control system is required, apart from the insulin signaling system. There are several lines of evidence suggesting that the AMPK signaling pathways can lead to the inhibition of PEPCK gene expression, though its mechanism is not fully understood.

In order to elucidate the molecular mechanism of AMPK observed in PEPCK gene expression, we searched for an unknown transcription factor, with the hypothesis that AMPK could mediate the transduction signal that leads to the repression of PEPCK gene expression through phosphorylation of unknown transcription factor.

Firstly, in order to identify responsive element(s) for AMPK in the human PEPCK gene promoter, functional analysis was performed using a series of promoter deletion mutants in a transient Luciferase reporter assay using primary rat hepatocytes. We found the DNA sequence which functioned as an AICAR dependent transcription repression. To identify the AICAR responsive element binding protein (referred to as AREBP), a yeast one-hybrid assay was carried out. Out of 10^8 colonies, three identical cDNA clones were isolated. There are several putative AMPK phosphorylation consensus sites in the AREBP molecule.

Next, we tested whether the isolated AREBP would actually act as an AICAR dependent regulator of the PEPCK gene by a transient Luciferase reporter assay using rat hepatoma cells. Although obvious differences were not observed with AREBP alone compared with mock transfection, the AREBP robustly repressed transcription in the presence of AICAR. In contrast, AREBP_{S470A} binding was not influenced by AMPK phosphorylation, even in the presence of AICAR. In addition, when the expression of endogenous AREBP was knocked down using the RNA interference (RNAi) technique, the repression of PEPCK expression following AICAR treatment was eliminated by AREBP-RNAi.

These results showed that AREBP would act as an AICAR dependent transcriptional repressor for PEPCK gene expression. It must be noted, however, that these results were derived from the analyses *in vitro* or at molecular level. In order to investigate the functions of AREBP *in vivo*, further researches are in progress using mouse with excessive AREBP expression. By understanding the regulation system of gluconeogenesis, we expect that our findings will be utilized for prevention and control of diabetes in the future.

AMP-activated protein kinase regulates PEPCK gene expression by direct phosphorylation of a novel zinc finger transcription factor.

Biochem Biophys Res Commun. 2006;351(4):793-9.

Inoue E and Yamauchi J

Nutritional Epidemiology Program, Bio-Index Project, National Institute of Health and Nutrition, Tokyo, Japan

Abstract: AMP-activated protein kinase (AMPK) acts as an intracellular sensor for maintaining the energy balance. Activation of AMPK switches on ATP-generating process while switches off ATP-consuming process. It achieves these effects by phosphorylation of downstream metabolic enzymes. It has been proposed that AMPK also regulates gene expression through phosphorylation of certain transcription factors; however its molecular mechanism is not fully understood. Here we show the cloning and characterization of a novel zinc finger transcription factor referred to as AREBP. AREBP is phosphorylated at Ser(470) by AMPK. Phosphorylation reduces the DNA-binding activity of AREBP. Transient transfection experiments indicate that wild-type AREBP, but not Ser(470) to Ala(470) substituted non-phosphorylating mutant, represses gene expression of the phosphoenolpyruvate carboxykinase (PEPCK), a key enzyme of gluconeogenesis. RNA interference-mediated reduction of endogenous AREBP expression attenuates AMPK-induced PEPCK down-regulation. These results implicate AREBP as a novel key modulator of PEPCK gene expression regulated by AMPK.

Latest Research

Young Japanese adults with high BMI tend to under-report energy intake – from the dietary records of the population aged 30's-60's.

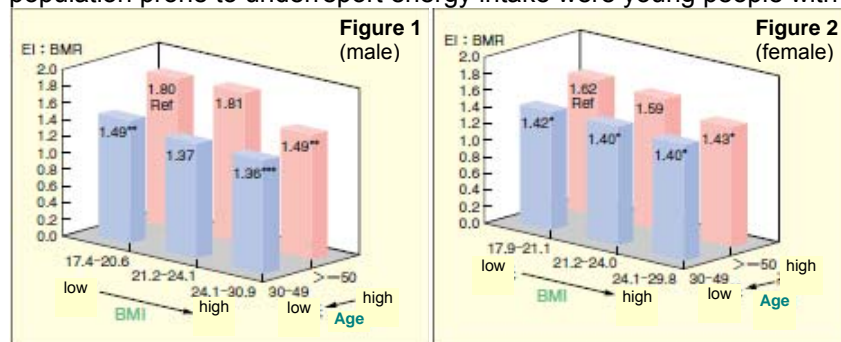
Hitomi Okubo[†], Satoshi Sasaki (Nutritional Epidemiology Program)

[†] Okubo H is currently with Kagawa Nutrition University

Reporting bias, underreporting or overreporting, is often observed in any type of self-reported dietary assessment methods. This is one of the big headaches for dietitians. So far, all the available studies on reporting bias were conducted in Western countries, and few studies has been undertaken in Japan. Diet record is one of the self-reported dietary assessment methods, which is believed to be reliable as the study subjects record what they actually took. The purpose of the present study was to examine the relative accuracy of self-reported energy intake obtained by diet records among various age ranges in the Japanese population.

We selected three areas which have different geographical conditions in Japan: Osaka (urban), Nagano (rural inland) and Tottori (rural coastal). Out of 96 married couples aged (30 years who were recruited, 183 subjects (91 women and 92 men) with complete 16-day diet records were included in the present analysis. The subjects completed 4-day semi-weighed diet records four times at 3-month intervals (16 days in total) from November 2002 to August 2003. We calculated the ratio of energy intake to basal metabolic rate (EI/BMR) to evaluate the relative accuracy of the reported energy intake. Low ratios describe subjects reporting comparatively low energy intake relative to their energy requirement, hence high likelihood of underreporting.

Mean EI/BMR was lowest in the subjects aged 30-39 years and highest in those aged 60 years or over (1.37 vs. 1.74 in male, 1.43 vs. 1.65 in female). A stepwise multiple regression analysis was performed to examine the prediction for relative accuracy of reporting. Regardless of sex, age correlated positively and BMI correlated negatively. **Figure 1 and 2** visually show the joint effect of age and BMI on EI/BMR values by cross-classifying subjects by both variables, with tertile of BMI on the x-axis, age group (30-49, ≥ 50 years) on the z and mean value of EI/BMR on the y. Compared with subjects classified into the lowest BMI and oldest age group, subjects in the highest BMI and youngest age group had significantly lower EI/BMR (**Fig 1**). Similar trend was observed in female as well (**Fig. 2**). These findings indicate that the characteristics of the population prone to underreport energy intake were young people with high BMI.



EI/BMR were adjusted for physical activity level and living area. * p<0.05, **p<0.01, ***p<0.001

Since the participants of this study might be highly health-conscious, we must be cautious to apply these trends to the whole general Japanese population. Yet, our findings would suggest that dietitians should bear in mind the possibility of underreporting when a nutritional assessment is undertaken for the subjects with high BMI or young ones.

The influence of age and body mass index on relative accuracy of energy intake among Japanese adults

Public Health Nutrition. 2006; 9(5): 651-657.

Okubo H¹, Sasaki S¹, Hirota N², Notsu A³, Todoriki H⁴, Miura A⁵, Fukui M⁶ and Date C⁷

1) Scientific Evaluation of Dietary Reference Intakes Project, National Institute of Health and Nutrition, Tokyo, Japan

2) Department of Living Sciences, Nagano Prefectural College, Nagano, Japan: 3) Tottori College, Tottori, Japan:

4) Department of Environmental and Preventive Medicine Faculty of Medicine, School of Medicine, University of

Ryukyus, Okinawa, Japan. 5) Department of Nutritional Health, Kwassui Women's College, Nagasaki, Japan:

6) Department of Statistics, Osaka City University Medical School, Osaka, Japan. 7) Department of Food Sciences and Nutrition, School of Human Environmental Sciences, Mukogawa Women's University, Hyogo, Japan

Abstract:

Objective: To examine relationships between the ratio of energy intake to basal metabolic rate (EI/BMR) and age and body mass index (BMI) among Japanese adults. **Design:** Energy intake was assessed by 4-day semi-weighed diet records in each of four seasons (16 days in total). The EI/BMR ratio was calculated from reported energy intake and estimated basal metabolic rate as an indicator of reporting accuracy. **Setting:** Residents in three areas in Japan, namely Osaka (urban), Nagano (rural inland) and Tottori (rural coastal). **Subjects:** One hundred and eighty-three healthy Japanese men and women aged ≥ 30 years. **Results:** The oldest age group (≥60 years) had higher EI/BMR values than the youngest age group (30-39 years) in both sexes (1.74 vs. 1.37 for men; 1.65 vs. 1.43 for women). In multiple regression analyses, age correlated positively (partial correlation coefficient, $\beta=0.012$, $P < 0.001$ for men; $\beta=0.011$, $P < 0.001$ for women) and BMI correlated negatively ($\beta=-0.031$, $P < 0.001$ for men; $\beta=-0.025$, $P < 0.01$ for women) with EI/BMR. **Conclusion:** Age and BMI may influence the relative accuracy of energy intake among Japanese adults.

Latest Research

Effects of age on ventilatory threshold and peak oxygen uptake normalized for regional skeletal muscle mass: a cross-sectional study on Japanese men and women aged 20-80 years.

Motohiko Miyachi (Health Promotion and Exercise Program)

【Background】 Ventilatory threshold (VT) is a valuable index of cardiorespiratory fitness as well as physical exercise intensity which is required for developing an exercise program for health promotion. However, few studies have examined the age-associated decline of VT. This study therefore aimed to demonstrate normative VT values and age-associated decline of VT normalized for skeletal muscle (SM) mass in 1,463 Japanese men and women aged 20-80 years.

【Methods】 Fourteen hundred and sixty-three healthy Japanese men and women, who are members of a fitness club in Tokyo, participated in this study (807 men and 656 women aged 20-80 years, with the mean age of 49.3 ± 13.5 years). SM mass was determined by B-mode ultrasound and estimated using the equations developed by Sanada. VT and peak oxygen uptake ($V_{O_{2peak}}$) were determined during treadmill walking, for which V_{O_2} were measured using an automated breath-by-breath mass spectrometry system. We developed the protocol in which the subjects warmed-up for 3 min, and then, the treadmill speed and grade were increased alternately for each successive minute of walking. $V_{O_{2peak}}$ was estimated using the heart rate (HR)- V_{O_2} relationship during sub-maximal exercise and the predicted maximal heart rates (HR)(220 minus age). VT was estimated from ventilatory equivalents for oxygen (VE/V_{O_2}) and carbon oxide (VE/V_{CO_2}). VT was determined as the point where V_{CO_2} increased sharply compared to V_{O_2} , or the one where VE/V_{CO_2} became unchanged and VE/V_{O_2} increased.

【Results and Discussion】 Total and regional (trunk and thigh) SM mass were negatively correlated with age in both men and women (Total SM mass decreased 0.13kg in men and 0.02kg in women per year, and the thigh SM mass decreased 0.052kg in men and 0.014kg in women per year). VT was significantly associated with thigh SM mass independent of age, body mass and fat-free mass. The most important finding in this study was that age-associated declines were observed in VT normalized for body mass in both men and women, but not in VT normalized for SM mass (Table). On the other hand, age associated declines were observed for $V_{O_{2peak}}$ normalized for body mass as well as normalized for SM mass. Thus, this study showed that the age-associated declines in VT are markedly blunted if normalized for SM mass rather than body mass. These results suggest that the decrease in VT with age is primarily due to an age-related decline of SM mass.

【Conclusion】 This study provided cardiorespiratory fitness data, using VT, according to gender and age group. Our findings would suggest that it is important to prevent the age-related decline in SM mass so as to prevent the age-associated declines in VT, namely cardiorespiratory fitness.

Table. Absolute and normalized VT in various age groups

Gender and age range (yrs)	n	Percentage of $V_{O_{2peak}}$ (%)	Absolute value (L)	Normalized values			
				Body mass (mL·kg ⁻¹ ·min ⁻¹)	Body mass ^{0.75} (mL·kg ^{-0.75} ·min ⁻¹)	Fat free body mass (mL·kg ⁻¹ ·min ⁻¹)	Thigh SM mass (mL·kg ⁻¹ ·min ⁻¹)
Men							
20-29	47	48.7 ± 7.8 †	1.71 ± 0.34 †	23.1 ± 4.2 †	97.5 ± 15.9 †	28.4 ± 4.9 †	162.4 ± 25.7
30-39	98	47.4 ± 8.1 †	1.48 ± 0.30 †	20.5 ± 3.6 †	85.6 ± 15.1 †	25.5 ± 4.5 †	151.7 ± 32.2
40-49	195	48.9 ± 7.4 †	1.47 ± 0.28 †	20.6 ± 3.3 †	85.5 ± 13.5 †	25.2 ± 4.2 †	154.6 ± 24.4
50-59	185	51.7 ± 8.2 †	1.40 ± 0.28 †	19.8 ± 3.3 †	81.6 ± 14.1 †	24.2 ± 4.0 †	153.0 ± 28.5
60-69	165	53.3 ± 9.3	1.26 ± 0.22 †	18.8 ± 3.2	76.1 ± 12.6	22.8 ± 3.8	148.2 ± 30.0
70+	65	58.0 ± 10.6	1.11 ± 0.19	17.4 ± 2.3	70.0 ± 9.7	20.9 ± 2.9	142.8 ± 22.9
All	755	51.1 ± 8.9	1.39 ± 0.31	19.9 ± 3.5	81.9 ± 15.0	24.3 ± 4.4	151.9 ± 28.1
Women							
20-29	47	51.3 ± 8.0	1.09 ± 0.20	20.5 ± 3.4 †	76.9 ± 13.0 †	27.2 ± 4.8 †	190.5 ± 30.9
30-39	144	50.4 ± 7.9	1.00 ± 0.22	19.9 ± 3.3 †	74.4 ± 12.8 †	25.9 ± 4.6 †	187.8 ± 35.2
40-49	161	54.5 ± 7.9	1.03 ± 0.20	19.4 ± 3.5	72.5 ± 13.2	25.7 ± 4.9 †	186.9 ± 39.1
50-59	148	55.4 ± 8.0	0.97 ± 0.18	18.4 ± 3.2	68.5 ± 12.1	24.1 ± 4.1	182.3 ± 32.9
60-69	100	58.7 ± 9.0	0.90 ± 0.16	16.8 ± 2.5	63.1 ± 9.6	22.7 ± 3.7	175.3 ± 36.1
70+	12	60.8 ± 9.9	0.86 ± 0.23	15.4 ± 3.2	55.6 ± 10.9	20.6 ± 4.1	180.8 ± 68.4
All	612	54.3 ± 8.6 *	1.00 ± 0.21 *	18.8 ± 3.5 *	70.5 ± 13.1 *	24.9 ± 4.7 *	183.9 ± 35.7 *

†, Significant difference in the 70-79-yr-old group ($P < 0.05$); *, Significant difference in all male subjects ($P < 0.05$).

Effects of age on ventilatory threshold and peak oxygen uptake normalised for regional skeletal muscle mass in Japanese men and women aged 20-80 years.

European Journal of Applied Physiology. 2007; 99:475-483

Sanada K¹⁾, Kuchiki T²⁾, Miyachi M³⁾, McGrath K⁴⁾, Higuchi M⁵⁾, Ebashi H⁶⁾,

1) Consolidated Research Institute for Advanced Science and Medical Care, Waseda University, Tokyo, Japan

2) Division of Integrated Humanistic and Cultural Studies, Graduate School of Integrated Science and Art, University of East Asia, Shimonoseki, Japan

3) National Institute of Health and Nutrition, Tokyo, Japan

4) Department of Physiological Sciences and Sports Performance, National Institute of Fitness and Sports, Kanoya, Japan

5) Faculty of Sport Sciences, Waseda University, Tokorozawa, Japan

6) Faculty of Integrated Cultures and Humanities, University of East Asia, Shimonoseki, Japan

Abstract: Ventilatory threshold (VT) is an important predictor of cardiorespiratory fitness, such as peak oxygen uptake ($V_{O_{2peak}}$) and is a valuable index of aerobic exercise intensity. However, little is known about the role of skeletal muscle (SM) mass in the age-associated decline of VT. Therefore, the present study was performed to investigate the effects of age on cardiopulmonary fitness normalised for regional SM mass in 1,463 Japanese men and women, and to determine the relevance of VT normalised to SM mass based on age and gender. Total, trunk and thigh SM mass were measured using an ultrasound method, while $V_{O_{2peak}}$ and VT were determined during treadmill walking. $V_{O_{2peak}}$ was estimated using the predicted maximum heart rate (HR) and the HR- V_{O_2} relationship for sub-maximal treadmill walking. There were significant negative correlations between VT normalised for body mass and age in men and women ($P < 0.001$). Age-associated declines were also observed in VT normalised for body mass in both men and women; however, VT normalised for SM mass was not significantly different with age. Significant correlations were also observed between thigh SM mass and VT in both men and women. These results suggest that thigh SM mass is closely associated with $V_{O_{2peak}}$ and/or VT in both men and women, and the decrease in VT with age is predominantly due to an age-related decline of SM mass. Moreover, this study provides normative cardiorespiratory fitness data regarding VT normalised SM mass in healthy men and women aged 20-80 years.