CHANGE OF COMPONENTS OF THE METABOLIC SYNDROME IN A WORKERS' HEALTH CHECKUP AFTER FIVE YEARS —RELATION WITH ELEVATED LIVER ENZYMES, GENE POLYMORPHISMS FOR ALDH 2, β3-AR AND LIFESTYLE

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We previously reported that the prevalence of elevated alanine aminotransferase (ALT) increases with accumulation of metabolic syndrome components, and a greater degree of involvement of aldehyde dehydrogenase 2 (ALDH2) than β3-adrenergic receptor gene (β3-AR) polymorphisms. The present study was designed to clarify the effect of aging, lifestyle and the two gene polymorphisms on the relationship between 4 components of the metabolic syndrome (obesity, hypertension, dyslipidemia and impaired glucose tolerance) and elevated ALT values in a subset of 73 out of 148 male workers who were 35 years of age in the baseline study and 40 years old in the present study.

Study subjects completed questionnaires about drinking and smoking habits, and underwent urinalysis, physical examination and peripheral blood tests, blood chemistry, electrocardiogram and chest X-rays each year as required by Japanese law. Information from the questionnaires and physical examinations, including liver function tests, were compared with previously reported ALDH2 and β3-AR genotypes for the 73 workers.

Of the 73 workers studied, 14 (19%) demonstrated decrease in metabolic syndrome components, 39 (53%) demonstrated no change, and 20 (27%) demonstrated an increase. Ten workers (14%) showed liver dysfunction at age 35 and 20 workers (27%) at age 40. Fourteen workers were newly diagnosed as having liver dysfunction at their 40-year checkup, thus being associated with the BMI and an active ALDH2 genotype. Accumulation of components of the metabolic syndrome were associated with the presence of liver dysfunction at 35 years.

In conclusion, these findings indicate that ALDH2 genotyping as well as lifestyle habits may be important factors in causing metabolic syndrome with liver dysfunction.

Key words: metabolic syndrome, alanine aminotransferase (ALT), ALDH 2, β3-AR

I. Introduction

Recent studies have provided evidence that nonalcoholic fatty liver disease (NAFLD) including nonalcoholic steatohepatitis (NASH) is a common manifestation of the metabolic syndrome[1~5], and the pathophysiology of both diseases seems to be largely attributable to insulin resistance. We reported that young male workers with elevated ALT might have NAFLD as an early manifestation of metabolic syndrome because they had two or more components of metabolic syndrome and the number of components increased with elevation of ALT. Moreover, persons with putative metabolic syndrome with elevated ALT demonstrated high body-mass-index (BMI), and serum triglycerides, as well as the active aldehyde dehydrogenase 2 (ALDH2) genotype[6]. An interesting finding in the baseline study was that 4 subjects with normal weight but putatively the metabolic syndrome with elevated ALT had both active ALDH2 and Arg64 β3-AR genotypes[6].

As our baseline study was cross-sectional, the present investigation was conducted to assess

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changes in metabolic syndrome components after an interval of 5 years in the same group of subjects[6]. Interventions, including treatment for the metabolic syndrome and education, especially the necessity of increased physical activity and improvement of diet, have been provided for the subjects over the past 5 years. We hypothesized that lifestyle is involved in development of the metabolic syndrome more than the ALDH2 and β3-AR genotypes. To test this hypothesis, gene polymorphisms of both ALDH2 and β3-AR and accumulation of components of metabolic syndrome were compared for 73 workers from the baseline study linked with new information for physical findings and lifestyle.

II. Subjects And Methods

Study Subjects

Of 148 car salesmen enrolled in the baseline study at 35 years[6], test results of 73 subjects (at age of 40) were available for the study. Twenty eight subjects were missing because of job change and laboratory data of 45 subjects were unavailable as they transferred to branch operations or subsidiary companies. We excluded two subjects who have been treated for lung cancer or drug induced hepatitis. The subjects for the baseline study were enrolled during September 1998 to August 1999 and the present study was performed from September 2003 to August 2004. During this period subjects were provided with health education by registered nurses, if necessary, with a focus on the importance of lifestyle in order to prevent lifestyle-related problems such as metabolic disease. The subjects were treated as needed with medications for hypertension, dyslipidemia, hyperglycemia, hyperuricemia and others by industrial physicians at the workplace, as well as by physicians at hospitals and/or clinics.

The industrial physician explained the purpose of the study to each worker. Each subject was asked to provide information on past medical history of illnesses and to fill out questionnaires to reveal drinking and smoking habits, diet, health conditions, physical activity, sleeping time and current drug use. The subjects underwent a physical examination, conventional laboratory tests including urinalysis, peripheral blood examination (red and white blood cell counts, hemoglobin), clinical chemistry (glucose, total cholesterol, triglyceride, HDL-cholesterol, aspartate aminotransferase (AST), alanine aminotransferase (ALT), γ-glutamyl transpeptidase (γ-GTP)), electrocardiogram, and chest X-rays at their annual health checkup. The ethics committee of the Tokai University School of Medicine as well as the Health Care Committee of the company approved the study protocol, and informed consent was obtained from each worker before participating in the study.

Alcohol Consumption and Smoking Habits

The subjects filled out questionnaires about alcohol consumption and smoking habits. To avoid underreporting the amount of alcohol or tobacco use and to obtain accurate data, we first explained to the subjects that the questionnaires would be used only for research purposes.

To assess drinking habits, the questionnaire asked for the number of days each week that alcohol was consumed, and how much alcohol was consumed on those days.

Questions on smoking habits asked for the number of cigarettes smoked per day and smoking history over the past 5 years.

Evaluation of Liver Dysfunction

The serum levels of three liver enzymes, i.e., AST, ALT, γ-GTP, were used to determine liver dysfunction. A subject was considered to have liver dysfunction with AST$>$40 IU/l, ALT$>$40 IU/l, and/or γ-GTP$>$60 IU/l.

Evaluation of Components of the Metabolic Syndrome

We evaluated the subjects for obesity, hypertension, dyslipidemia and impaired glucose tolerance as the four key components of the metabolic syndrome. We categorized the subjects into the following three groups according to BMI criteria established by the Japan Society for the Study of Obesity (JASSO): lean (BMI$<$18.5), normal (18.5$\leq$BMI$<$25.0), and obese (25.0$\leq$BMI). Hypertension (systolic blood pressure$\geq$130 mmHg and/or diastolic blood pressure$\geq$80 mmHg), dyslipidemia (total cholesterol level$\geq$220 mg/dl and/or triglyceride level$\geq$150 mg/dl), and impairment of glucose tolerance (fasting plasma glucose level$\geq$110 mg/dl) were also assessed.

Gene Polymorphisms for ALDH 2 and β-AR

The methodology for determination of both genes was reported previously[6]. The genotyping data from the baseline study were linked to the new information in this study. Subjects with the ALDH2*1/*1 genotype were classified as having active ALDH2, and subjects with the ALDH2*1/*2 or ALDH2*2/*2 genotype were classified as having inactive ALDH2. The Arg64Arg and Trp64Arg polymorphism of the β3-AR gene were classified as the Arg64 genotype and the Trp64Trp genotype as the Trp64 genotype.

Statistical Analyses

Categorical variables were assessed by the chi-square or Fisher's exact tests. Quantitative values are expressed as means±standard deviations (SDs). The significance of differences between groups was

analyzed by ANOVA, then a post hoc test (Sheffe's test). Logistic regression analysis was performed to predict factors that are associated with liver dysfunction 5 years later or accumulation of components of the metabolic syndrome. A level of $p < 0.05$ was considered significant. All analyses were performed with the computer program StatView 5.0 (SAS Institute Inc., Cary, NC) except for the logistic regression analysis, which was performed with SPSS 12.0 (SPSS Inc., Tokyo, Japan).

III. Results

Characteristics of the 73 Male Subjects

The physical and clinical chemistry data from the 73 male workers at age 35 and age 40 were compared (Table 1). None of the subjects had chronic liver disease caused by hepatitis B or C viral infection, alcohol, drugs, or autoimmune hepatitis. In the baseline study, 28 of 148 (19.0%) subjects had liver dysfunction according to the criteria described in the Subjects and Methods. Of the 73 subjects studied in this study, 10 (14%) had liver dysfunction at age 35 and 20 (27%) at age 40.

Of the 148 subjects who were age 35 in the baseline study, 8 (5.4%) had elevated AST levels, 23 (15.5%) had elevated ALT levels, and 14 (9.5%) had elevated γ-GTP levels. In the 73 subjects studied at both age 35 and age 40, AST levels were elevated in 2 (2.7%) at 35 years and 4 (5.4%) at 40 years, ALT levels were elevated in 9 (12.3%) at 35 years and 14 (19.2%) subjects at 40 years, and γ-GTP levels were elevated in 6 (8.2%) at 35 years and 13 (17.8%) at 40 years, respectively. The proportion of subjects with each liver enzyme elevated increased over the 5 year period. Patient characteristics at age 35 in the present study ($n = 73$) were otherwise basically similar to those at age 35 in the baseline study ($n = 148$).

Subjects with elevated liver enzymes underwent ultrasonography to confirm fatty infiltration of the liver. Excluding other liver diseases by serological examination and the questionnaire regarding drinking habits (less than 30 g of alcohol per day), a majority of subjects were diagnosed as having nonalcoholic fatty liver disease (NAFLD).

The percentages of subjects who had obesity, hypertension, dyslipidemia, or impaired glucose tolerance were 28.8%, 58.9%, 41.1%, and 5.5% at age 35 and 39.7%, 57.5%, 41.1%, and 11.0% at 40, respectively (Table 1). There was only one subject with elevated liver enzymes without any of the four components of the metabolic syndrome at age 40.

Comparison of Liver Function Among Four Groups Classified According to the Number of Components of Metabolic Syndrome at Ages 35 and 40

Seventeen (23.3%) of the 73 subjects had no components of metabolic syndrome, 21 (28.8%) had one component, 24 (32.9%) had two, 11 (15.1%) had more than three at age 35 (Table 2-a). Fifty-six subjects (76.7%) had one or more components of metabolic syndrome at age 35. At age

Table 1. Characteristics of the 73 male workers in the present follow-up study

	Age 35	Age 40
Age (yr)	35.2±0.4	39.3±0.5
Height (cm)	171.4±6.7	171.3±6.1
Body weight (kg)	69.3±11	70.7±11.2
Body mass index (kg/m^2)	23.6±3.4	24±3.5
Systolic blood pressure (mmHg)	128±15.3	128.4±13.3
Diastolic blood pressure (mmHg)	81.1±11.2	79±9.4
AST (IU/L)	21.3±7.9	23.1±8.2
ALT (IU/L)	26.4±19.2	28.9±17.9
γ-GTP (IU/L)	28.5±23.8	45.4±30.8
Fasting plasma glucose (mg/dL)	90.7±9.1	97±15.2
Total cholesterol (mg/dL)	210.9±43.5	206.6±36.8
HDL-cholesterol (mg/dL)	59.7±11.5	59±11.3
Triglyceride (mg/dL)	106.7±62.2	115.6±57.6
No. of Subjects (%)		
Obesity*	21(28.8%)	29(39.7%)
Hypertension†	43(58.9%)	42(57.5%)
Dyslipidemia‡	30(41.1%)	30(41.1%)
Impaired glucose tolerance§	4(5.5%)	8(11.0%)
Smoking¶		
Never smoking	22(30.1%)	22(30.1%)
Former smoking	6(8.2%)	6(8.2%)
Current smoking		
1–19	12(16.4%)	12(16.4%)
≥20	33(45.2%)	33(45.2%)
Drinking¶		
Never	15(20.5%)	15(20.5%)
<30 g/day	34(46.6%)	34(46.6%)
≥30 g/day	24(32.9%)	24(32.9%)

* BMI of ≥25.0, according to the standard Japanese diagnosis of obesity.

† Systolic blood pressure ≥130 mm Hg and/or diastolic blood pressure ≥80 mm Hg.

‡ Triglyceride level ≥150 mg/dl and/or HDL-cholesterol ≥40 mg/dL.

§ Fasting plasma glucose level ≥110 mg/dl.

¶ Smoking and drinking habits at 40 years were the same as those at 35 years.

Table 2-a. Accumulation of components of the metabolic syndrome and the levels of AST, ALT, γ-GTP at age 35.

	Number of Components				
	0	1	2	≥3	
No. of Subjects	17	21	24	11	*P* value
AST (IU/L)	19.5±3.8	18.5±5.7	23.4±9.7	27.3±8.4*	0.007
ALT (IU/L)	18.2±6.7	17±7.1	32.9±23.7†	42.6±22.1‡	<0.001
γ-GTP (IU/L)	17.8±8.1	18.4±7.5	36.5±25.0	46.6±38.8¶	<0.001

* $P=0.021$ v subjects having 1 component by Scheffe's test.
† $P=0.002$ v subjects having no component by Scheffe's test.
‡ $P=0.001$ v subjects having 1 component and $P=0.005$ v subjects having no component by Scheffe's test.
¶ $P=0.010$ v subjects having no component and $P=0.009$ v subjects having 1 component by Scheffe's test.

Table 2-b. Accumulation of components and levels of AST, ALT, γ-GTP at age 40

	Number of Components				
	0	1	2	≥3	
No. of Subjects	15	26	17	15	*P* value
AST (IU/L)	20.1±9.9	21.5±6.3	32.2±5.9	29.9±8.6*	0.002
ALT (IU/L)	18.1±9.7	23.6±10	31.7±19.5	45.6±21.1†	<0.001
γ-GTP (IU/L)	29.6±13.7	36±16.1	51.3±37.6‡	70.6±38‡	<0.001

* $P=0.008$, 0.01, and 0.008 v subjects with no component, subjects with 1 component, and subjects having 2 components, respectively by Scheffe's test.
† $P=<0.0001$ v subjects with non component and subjects with 1 component by Scheffe's test.
‡ $P=0.001$ and 0.003 v subjects with no component and subjects with 1 component by Scheffe's test

40, 26 (35.6%) had one component, 17 (23.3%) had two components, and 15 (20.5%) had more than three (Table 2-b). The values of AST and ALT were similar in both age groups, but γ-GTP levels increased after 5 years even in workers with no components. The AST, ALT and γ-GTP levels increased with the number of components of metabolic syndrome in both age groups (at age 35; AST, $P=0.007$; ALT, $P=0.0001$; γ-GTP, $P=0.0005$; at age 40, AST, $P=0.023$; ALT, $P<0.0001$; γ-GTP, $P=0.0003$).

Association of Liver Dysfunction with Frequency of the ALDH2 and β3-AR Genotypes

Table 3 shows the association of liver dysfunction with the ALDH2 and β3-AR genotypes. Active ALDH2 was seen more frequently among those with liver dysfunction than among those who did not ($P=0.046$, chi-square test), although this relationship was not significant in the baseline study ($P=0.0745$). No relationship between the β3-AR gene polymorphism and liver function was evident among the subjects in either the baseline or present studies.

Association of Liver Dysfunction with Frequency of ALDH2 and β3-AR Genotypes Among Subjects with Normal BMI

In the normal BMI group, persons with elevated ALT (>40) were more likely to have active ALDH2 and the Arg64 genotype of β3-AR ($18.5\leq$ BMI<25.0) in the baseline study. In the lean and obese BMI groups, there was no correlation between liver dysfunction and prevalence of the β3-AR or

Table 3. Prevalence of ALDH2 and β3-AR genotypes in subjects with or without liver dysfunction

	Subjects With Liver Dysfunction (n=20)	Subjects Without Liver Dysfunction (n=53)	
ALDH2 genotype			
ALDH2*1/*1	15(75.0%)	26(49.1%)	
ALDH2*1/*2	4(20.0%)	24(45.3%)	
ALDH2*2/*2	1(5.0%)	3(5.7%)	
ALDH2 activity			
Active (ALDH2*1/*1)	15(75.0%)	26(49.1%)	$\chi^2=3.97$
Inactive (ALDH2*1/*2 +ALDH2*2/*2)	5(25.0%)	27(50.9%)	$P=0.046$
β3-AR genotype			
Trp64Trp	15(75.0%)	43(81.1%)	
Trp64Arg	4(20.0%)	9(17.0%)	
Arg64Arg	1(5.0%)	1(1.9%)	
β3-AR allele			
Trp64	15(75.0%)	43(81.1%)	$\chi^2=0.33$
Arg64	5(25.0%)	10(18.9%)	$P=0.563$

ALDH2 genotypes. In the present study, all 4 subjects with elevated ALT without obesity had the active ALDH2 genotype (Table 4).

Association of ALT Levels with the ALDH2 and β3-AR Genotypes in Subjects of the Normal BMI Group (18.5≤ BMI<25.0)

We analyzed the relationship between ALT levels and the ALDH2 or β3-AR genotype in the normal BMI group (18.5≤BMI<25.0) by two-way ANOVA and post hoc test in both studies. The ALT levels of subjects with the Arg/Arg or Trp/Arg genotype of β3-AR were significantly higher than those of subjects with the Trp/Trp genotype of β3-AR in the baseline study, but not higher in the present study ($P=0.0390$ in the baseline study vs. $P=0.3585$ in the present, Scheffe's test).

The ALT level of subjects with active ALDH2 was also higher than that of subjects with inactive ALDH2 ($P=0.0286$ in the baseline study vs. $P=0.0592$ in the present, Scheffe's test).

The AST level was significantly higher among those with the Arg/Arg or Trp/Arg genotype of β3-AR compared to those with the Trp/Trp genotype in the baseline study but not in the present study.

Association of Alcohol Drinking, Smoking Habits, and Appearance of Liver Dysfunction with Number of Metabolic Syndrome Components

As in the baseline study, there were no significant differences in the amount of daily alcohol intake and smoking habit between those who did and those who did not have liver dysfunction. Of the 73 workers studied, 14 (19%) demonstrated decrease in metabolic syndrome components, 39 (53%) demonstrated no change, and 20 (27%) demonstrated an increase. There were neither significant differences in alcohol drinking nor in smoking habits among the three groups.

Fourteen workers were newly diagnosed as having liver dysfunction at their 40-year checkup. To select the factors significantly associated with developing liver dysfunction in these 14, a logistic regression analysis was performed. Drinking and smoking habits, ALDH2 and β3-AR genotypes (categorical variables), and BMI (continuous variables) at 40 years were chosen as independent variables. By forward stepwise variable selection, the BMI at 40 years and the active ALDH2 genotype were independently associated with development of liver dysfunction at 40 years (Table 5).

In the present study 15 out of 73 workers had more than 3 components of the metabolic syndrome. To clarify the factors associate with accumulation of components of the metabolic syndrome, logistic regression analysis was performed with variables including smoking and drinking habits at 40 years, ALDH2 and β3-AR genotypes, and presence of liver dysfunction at 35 years (categorical variables). Among these factors liver dysfunction at 35 years was significantly associated with accumulation of metabolic syndrome components (Table 6).

Table 5. Factors associated with development of liver dysfunction* by logistic regression analysis

Independent variables	Odds ratio	95% confident interval	*P* value
BMI	11.88	2.56–55.16	0.002
ALDH2 (active)	5.44	1.10–26.90	0.038

* Fourteen workers were newly diagnosed as having liver dysfunction in the present study. The checkup data of these 14 workers were compared with those for 49 workers who did not have liver dysfunction in the present study.

Table 4. Relationship between elevated ALT and the ALDH2 and β3-AR genotypes

	ALDH2 genotype				
	Active		Inactive		
β3-AR genotype	Arg64Arg Trp64Arg	Trp64Trp	Arg64Arg Trp64Arg	Trp64Trp	
All subjects (n=73)					
ALT>40 IU/L (n=14)	2(14.2%)	9(64.3%)	1(7.1%)	2(14.3%)	
ALT≤40 IU/L (n=59)	7(11.9%)	23(39.0%)	5(8.5%)	24(40.7%)	$P=0.224$*
18.5≤BMI<25 (n=41)					
ALT>40 IU/L (n= 4)	1(25.0%)	3(75.0%)	0(0.0%)	0(0.0%)	
ALT≤40 IU/L (n=37)	6(14.6%)	13(31.7%)	4(9.8%)	14(14.0%)	$P=0.308$*

* Frequency analysis was performed with an extended Fisher's exact test.

Table 6. Factors associated with accumulation of the metabolic syndrome components*

Independent variables	Odds ratio	95%CI‡	*P* value
Smoking			
Never smoking§	1.00		
Former smoking	1.32	0.27–6.52	0.732
1–19	0.14	0.01–3.63	0.236
20≤	0.95	0.13–7.06	0.959
Drinking	4.06	0.35–46.23	0.258
Never drinking¶	1.00		
<30 g/day	0.21	0.02–2.67	0.227
≥330 g/day	0.91	0.19–4.29	0.904
β3-AR (Arg64 genotype)	9.16	0.57–145.51	0.116
ALDH2 (active)	1.11	0.19–6.40	0.906
Liver dysfunction†	24.69	2.47–246.77	0.006

* Subjects having 3 or more components of the metabolic syndrome in the present study. Forced entry: All variables entered in a single step.
† Liver dysfunction at the baseline.
‡ Confidence interval.
§ Never smoking used as the reference.
¶ Never drinking used as the reference.

IV. Discussion

A close association of liver dysfunction and the metabolic syndrome, related to a polymorphism in the ALDH2 gene was demonstrated in both the present and baseline studies[6], although the numbers of subjects did not provide sufficient statistical evidence for unequivocal confirmation. At the beginning of the present study, we hypothesized that ALDH2 and β3-AR genotypes are involved in the development of metabolic syndrome in younger persons, especially normal weight persons, and that accumulation of unhealthy habits may affect its occurrence in the middle and older aged persons because it has been demonstrated that the prevalence of metabolic syndrome increases with age[7,8]. In order to clarify this hypothesis we enrolled 148 workers in this study when they were 35 years old, as a blood test was performed at this age for the first time after joining the company, and 73 of 148 workers could be followed up after 5 years. Among them, a total of 20 workers had liver dysfunction, and 4 of them showed putative metabolic syndrome with elevated ALT with normal weight. Contrary to our baseline study[6], all 4 workers had active ALDH2 but not the Arg64 genotype of β3-AR gene. The results indicate that the β3-AR gene polymorphism is not involved in development of the metabolic syndrome with elevated ALT in middle-aged persons.

Workers with elevated liver enzymes who had no viral markers or other causes such as alcohol, drug, or autoimmune mechanisms, exhibited increased numbers of metabolic syndrome components (abdominal obesity, dyslipidemia, hypertension, and glucose intolerance) in both the present and baseline study[6]. As mentioned previously, the deposition of fat in the abdominal fat cells may cause flux of free fatty acid to the liver, resulting in the development of fatty liver[1~5]. Therefore, one might expect to observe elevated liver enzymes at the beginning of the metabolic syndrome, though conclusive evidence was not obtained in the present study. We are still trying to confirm this theory, because early detection of metabolic syndrome in young adults using liver enzymes would be very useful. In fact, insulin resistance evaluated by HOMA-IR was present in younger adults in a cross-sectional study in Spain[8]. They suggested that these young persons with insulin resistance may progress to metabolic syndrome in middle or older age. ALT might replace HOMA-IR to predict the occurrence of metabolic syndrome.

"Syndrome X" by Reaven[9], "Deadly Quartet" by Kaplan[10], "Insulin Resistance Syndrome" by DeFronzo[11], "Visceral Obesity Syndrome" by Matsuzawa[12], "Metabolic Syndrome X" by Matsuzawa[13], "Multiple Metabolic Syndrome" by Liese[14], "Multiple Risk Factor Syndrome"[15] have a common pathophysiology, that is insulin resistance, and are now collectively called metabolic syndrome. A WHO working group established a definition and diagnostic criteria for the disease[16]. The Third National Health and Nutrition Examination Survey (NHANES III, Adult Treatment Panel III report) reported new criteria defining metabolic syndrome to consist of more than 3 of the following 5 risk factors: (1) waist circumference ≧102 cm in men and ≧88 cm in women, (2) TG ≧150 mg/dl, (3) HDL-C ≦40 mg/dl, (4) arterial blood pressure ≧130/85 mmHg, and (5) fasting blood sugar ≧110 mg/dl[7]. Very recently, Matsuzawa announced the following new diagnostic criteria for metabolic syndrome for Japanese on behalf of eight Japanese academic societies[17]. A waist circumference of ≧85 cm in men and ≧90 cm in women is necessary for the diagnosis, and the two of the following three conditions should be present[17]. The Adult Treatment Panel III report as well as the new criteria for Japanese emphasized the presence of abdominal obesity among other risks. As periodic health checks regulated by law (the Community Health Law and the Labor Safety and Health Law) in Japan do not require the measurement of waist circumference, we here used BMI instead of waist circumference. If waist circumference

were available, a relation between the increase in the number of the metabolic syndrome components and/or elevated ALT and gene polymorphism might be observed. A relationship between the ALDH2 gene polymorphism and the occurrence of metabolic syndrome with elevated liver enzymes was seen among normal BMI persons in the baseline study as well as in the present study.

The β3-AR gene polymorphism is thought to be related to abdominal obesity and insulin resistance[18~22]. Several reports have described a relationship between fatty liver and insulin resistance[2]. Therefore, β3-AR gene polymorphism may be involved in the development of fatty liver in the early phase. Though few reports have described relationships between obesity and the β3-AR gene polymorphism, difficulty in losing weight in people with the Arg64 genotype has been descrived[23,24]. We selected the ALDH2 gene as another candidate gene for fatty liver because the ALDH2 genotype is related to the alcohol drinking habit[25~27]. As observed in the baseline study[6] the present study showed a high prevalence of active ALDH2 among workers with elevated ALT. Our results are similar to those of a study that revealed that habitual drinkers with active ALDH2 showed liver dysfunction more often than subjects with inactive ALDH2[28]. ALDH2 may participate in the metabolism of not only alcohol but also other chemicals with aldehyde radicals, resulting in alteration of lipid metabolism as mentioned previously[6].

Effects of smoking and/or drinking alcohol on prevalence of the metabolic syndrome, especially on the dynamic condition of insulin resistance, have been reported[29~34]. We could not confirm any influence of either alcohol consumption or smoking on the development of metabolic syndrome in the present study. The reason may be that the number of subjects was too small. Another is that we used relatively young persons as subjects, and it takes time to accumulate components of the metabolic syndrome. The present study did show that liver dysfunction at 35 years was associated with accumulation of components of the metabolic syndrome at 40, indicating that liver dysfunction is an early symptom. Other lifestyle related factors such as diet and physical activity might be involved. The present study lacks information concerning these lifestyle related factors. Increased physical activity should be evaluated for the treatment of metabolic syndrome with liver dysfunction and ALDH2 gene polymorphism, though health education is very difficult[35].

Most reports on metabolic syndrome have been cross-sectional studies[36], and there have been no prospective studies to date. A longitudinal study is necessary to clarify the involvement of genetic factors in development of the metabolic syndrome, and temporal relationship with fatty liver. Therefore, we will continue to follow-up workers and accumulate health data in order to develop prevention guidelines for metabolic syndrome based on the impact of gene polymorphism.

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