Young Men With High-Normal Blood Pressure Have Lower Serum Adiponectin, Smaller LDL Size, and Higher Elevated Heart Rate Than Those With Optimal Blood Pressure

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OBJECTIVE — Three measures—heart rate, a global index of the influence of the autonomic nervous system on the heart; circulating concentrations of adiponectin, an adipose-specific protein; and C-reactive protein (CRP), a sensitive marker of inflammation—have been reported to be closely associated with insulin resistance. Patients with borderline hypertension are known to be more insulin resistant and dyslipidemic than those with normal blood pressure (BP). BP can be classified into three categories: optimal, normal, and high-normal. The present study examined whether those with high-normal BP have any of these three conditions as compared with those with optimal BP in young healthy men.

RESEARCH DESIGN AND METHODS — Anthropometric, blood pressure, heart rate, and blood tests, including tests for adiponectin and CRP, were conducted in 198 male students, ages 18–26 years, who had fasted overnight. Insulin resistance (IR) and insulin secretion (β-cell levels) were calculated using the homeostasis model assessment (HOMA), and LDL size was measured by PAGE.

RESULTS — Compared with the 90 men who had optimal BP, the 46 men with high-normal BP had increased heart rate, BMI, percent body fat, and serum leptin levels. In addition, they had greater serum insulin, HOMA IR, and β-cell levels, lower adiponectin levels, and comparable CRP levels. Furthermore, the 46 men with high-normal BP had higher serum triglyceride and apolipoprotein (apo) B levels, and smaller LDL size; however, there was no difference in LDL and HDL cholesterol and apoA-I between men with optimal and high-normal BP. After adjusting for BMI, differences were still significant in serum adiponectin, heart rate, and LDL particle size. As BP rose, there was an increase in heart rate (BMI-adjusted least square means were 63, 65, and 70 bpm in men with optimal, normal, and high-normal BP, respectively; $P = 0.005$), whereas serum adiponectin (7.5, 6.6, and 6.4 mg/l; $P = 0.007$) and LDL particle size (271, 269, and 269 Å; $P = 0.008$) decreased.

CONCLUSIONS — Young men with high-normal BP have a faster heart rate, lower serum adiponectin levels, and smaller LDL size than men with optimal BP, even after adjustment for BMI. These results suggest the necessity of preventing further development of cardiac and metabolic diseases in young people who have high-normal BP.

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Abnormalities of glucose, insulin, and lipoprotein metabolism are common in patients with hypertension (1,2). These changes can also be discerned in borderline hypertensive patients (3,4). Recently, normal blood pressure (BP) has been classified into three categories: high-normal, normal, and optimal (5). High-normal BP has been shown to be a strong predictor of type 2 diabetes (6), similar to hypertension (7). The aim of the present study was to determine whether abnormalities of glucose, insulin, and lipoprotein metabolism are more common in individuals with high-normal BP than in those with optimal BP. To do this, we examined a homogeneous cohort of young healthy Japanese men, only 5% of whom were found to have high BP (8). In addition, we measured heart rate, a global index of the autonomic nervous system influence on the heart; serum concentrations of C-reactive protein (CRP), a sensitive marker of inflammation; and adiponectin, an adipose-specific protein, all of which have recently been reported to be associated with insulin resistance (9–11).

RESEARCH DESIGN AND METHODS

— In 1997, 198 men entered Kobe University of Merchantile Marine, Kobe, for this study. All were of Japanese nationality, and 190 (96%) of them were ages 18–20 years. In all, 179 serum samples were available for CRP and adiponectin measurements (Table 1); there were no significant differences between the 179 men and the remaining 19 men for whom sera were not available (data not shown).

Students were asked to fast overnight

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Abbreviations: apo, apolipoprotein; BP, blood pressure; CRP, C-reactive protein; DBP, diastolic BP; FI, fasting insulin; FPG, fasting plasma glucose; HOMA, homeostasis model assessment; IR, insulin resistance; SBP, systolic BP.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

and to refrain from smoking and ingesting alcohol overnight before attending the center. Body weight and percent body fat were measured after they voided. This was done using an impedance fat meter (TBF-202; Tanita Corp., Tokyo, Japan). TBF-202 uses two foot-pad electrodes with a corresponding digital scale, as previously reported (8).

Alcohol consumption and smoking habits were determined by an interview at the time of each participant's physical examination. Data with respect to diet and exercise were not available. No subject was currently receiving any medications.

BP was measured with a standard mercury sphygmomanometer after the subjects had rested at least 10 min. Systolic BP (SBP) was recorded at the appearance of sounds, and diastolic BP (DBP) was recorded at the disappearance of sounds (V-phase Korotkov). The measurements were repeated after 2–3 min, and the average of the measurements was used for analysis.

Electrocardiograms (12 lead) were recorded at a 25 mm/s paper speed and at 10 mm/mV gain by means of an automated electrocardiogram (FCP-4,266; Fukuda Denshi, Tokyo, Japan). The computer program uses successive R-R intervals between all ventricular muscle depolarizations (QRS complexes) to calculate the mean heart rate within the recorded period of 10 s. Sinus rhythm was present in all participants, and repolarization disturbances were not detected in any electrocardiogram.

Hypertension was defined as a SBP ≥140 mmHg and/or a DBP ≥90 mmHg, high-normal BP as a SBP = 130–139 mmHg or a DBP = 85–89 mmHg, normal BP as a SBP = 120–129 mmHg and a DBP = 80–84 mmHg, and optimal BP as a SBP <120 mmHg and a DBP <80 mmHg (5). When SBP and DBP fell into different categories, the higher category was used. As previously reported (8), high BP was found in only 10 (5%) of the 198 men studied, optimal and normal BP in 90 (46%) and 52 (26%) men, respectively, and high-normal BP in 46 (23%) men.

Venous blood was sampled after an overnight fast and centrifuged at 3,000 rpm for 30 min at 4°C. Plasma glucose was measured by the glucose oxidase method. Insulin and leptin were assayed using commercially available kits (Pharmacia, Tokyo, Japan, and Linco Research, St. Charles, MO, respectively). HDL cholesterol levels were measured after precipitation of LDL and VLDL particles with dextran sulfate magnesium (12).

Table 1—*Anthropometric and biochemical characteristics of 198 young male college students*

Variables	Means ± SD
SBP (mmHg)	121 ± 12
DBP (mmHg)	71 ± 8
Heart rate (bpm)	64 ± 10
BMI (kg/m^2)	21.6 ± 3.6
Body fat (%)	18.3 ± 5.9
Leptin (ng/ml)	2.1 ± 2.7
Fasting plasma glucose (mmol/l)	4.95 ± 0.41
Fasting insulin (pmol/l)	49 ± 20
HOMA IR	1.83 ± 0.82
HOMA β-cell	119 ± 54
CRP (mg/l)	0.51 ± 1.45
Adiponectin (mg/l)	6.9 ± 2.8
Total cholesterol (mmol/l)	4.56 ± 0.76
LDL cholesterol (mmol/l)	2.75 ± 0.69
HDL cholesterol (mmol/l)	1.50 ± 0.25
Triglycerides (mmol/l)	0.70 ± 0.36
ApoA-I (g/l)	1.33 ± 0.18
ApoB (g/l)	0.73 ± 0.19
LDL size (Å)	270 ± 5

Cholesterol (13) and triglyceride (14) levels were measured by the respective enzymatic method using a Hitachi 7170 autoanalyzer (Mito, Ibaragi, Japan). LDL cholesterol was calculated using the formula of Friedewald et al. (15).

The diameter of the major LDL fraction was determined by gradient gel electrophoresis on 2–16% polyacrylamide gels (Biocraft, Tokyo, Japan) according to the method of Nichols et al. (16). Plasma and three standards of known diameter—apoferritin (12.2 nm), thyrogloblin (17.0 nm), and latex beads (39.0 nm)—were simultaneously electrophoresed on the same slab gel for 24 h. After electrophoresis, LDL lipid was stained with Oil Red O, and the standards were stained with Coomassie G-250. The average LDL particle diameter was estimated by interpolation from a plot of the logarithm of the diameter of the standards versus the migration distance of the standards.

Insulin resistance (IR) and secretion (β-cell) determined by homeostasis model assessment (HOMA) (17) were calculated using fasting plasma glucose (FPG) and fasting insulin (FI) levels in each participant, as follows:

HOMA IR = FPG (mmol/l) × FI (μU/ml)/22.5

HOMA β-cell = 20 × FI (μU/ml)/(FPG [mmol/l] – 3.5)

HOMA IR has been validated by comparison with results of glucose clamp studies (17,18), intravenous glucose tolerance test (17,19), and continuous infusion of glucose with minimal model assessment (19). The HOMA β-cell method has been validated by comparison with the intravenous glucose model assessment (20). Application of HOMA has also been used in epidemiological studies (17,21,22).

CRP and adiponectin were measured in sera stored at –70°C using a highly sensitive immunonephelometric assay (23) and an enzyme-linked immunosorbent assay (24), respectively. The intra- and interassay coefficients of variations of adiponectin were 3.3 and 7.4%, respectively, and those of CRP were <5%.

The SAS statistical program (SAS Institute, Cary, NC) was used to perform all statistical analyses. Significance of differences in mean values across BP status was assessed by two-way ANOVA and then ANCOVA, followed by Tukey-Kramer's procedure (25). $P < 0.05$ was considered significant. Data are expressed as mean ± SD unless otherwise stated.

RESULTS — As previously reported (8,26,27) and confirmed in the present study (Table 1), most young Japanese men studied were normotensive, nonobese, and nondiabetic and had high HDL cholesterol, low triglyceride, and desirable LDL cholesterol levels. In addition, they were not hyperinsulinemic, and their CRP levels averaged 0.51 mg/l. Furthermore, they had large LDL size, with a mean of 270 Å.

As compared with men with optimal BP, those with high-normal BP had a faster heart rate (Table 2) and higher BMI, percent body fat, and serum leptin levels. Furthermore, they had elevated serum insulin concentrations, HOMA β-cell, HOMA IR, and lower serum adiponectin, but had comparable serum CRP levels. Finally, men with high-normal BP had higher serum levels of triglycerides and apolipoprotein (apo)-B and smaller LDL size. However, there were no significant differences in total, LDL or HDL cholesterol, and apoA-I between the two groups.

Men with normal BP had results that were intermediate between those in the

Table 2—*Blood pressure, anthropometry, and biochemical data in young college men stratified by blood pressure status*

	Blood pressure status				*P*	
	Optimal	Normal	High-normal	High	Unadjusted	Adjusted for BMI
n	90	52	46	10	—	—
SBP (mmHg)	110 ± 7	124 ± 3	133 ± 4	144 ± 6	0.0000	0.00
DBP (mmHg)	65 ± 6	73 ± 5	79 ± 5	83 ± 7	0.0000	0.00
Heart rate (bpm)	63 ± 12	65 ± 9	70 ± 10	67 ± 9	0.0055	0.007
BMI (kg/m^2)	20.4 ± 2.2	21.4 ± 2.2	23.1 ± 4.8	26.5 ± 6.5	0.0000	—
Body fat (%)	16.1 ± 3.7	18.2 ± 3.9	20.9 ± 7.7	25.6 ± 8.6	0.0000	0.15
Leptin (ng/ml)	1.5 ± 0.7	1.7 ± 0.7	3.2 ± 4.5	5.9 ± 5.2	0.0000	0.18
Plasma glucose (mmol/l)	4.89 ± 0.44	5.06 ± 0.33	5.06 ± 0.39	4.94 ± 0.50	0.0658	0.11
Insulin (pmol/l)	43 ± 12	50 ± 12	57 ± 30	68 ± 29	0.0002	0.14
HOMA IR	1.56 ± 0.49	1.90 ± 0.58	2.12 ± 1.16	2.55 ± 1.31	0.0000	0.14
HOMA β-cell	113 ± 40	112 ± 31	129 ± 82	168 ± 72	0.0055	0.17
CRP (mg/l)	0.32 ± 0.55	0.76 ± 2.50	0.42 ± 0.72	0.92 ± 0.96	0.2156	0.45
Adiponectin (mg/l)	7.9 ± 3.0	6.5 ± 2.4	6.4 ± 2.4	5.6 ± 2.7	0.0017	0.002
Total cholesterol (mmol/l)	4.45 ± 0.70	4.65 ± 0.75	4.60 ± 0.70	5.02 ± 1.24	0.0610	0.34
LDL cholesterol (mmol/l)	2.64 ± 0.65	2.82 ± 0.67	2.77 ± 0.70	3.21 ± 0.98	0.0557	0.43
HDL cholesterol (mmol/l)	1.53 ± 0.23	1.53 ± 0.26	1.47 ± 0.26	1.32 ± 0.21	0.0641	0.67
Triglycerides (mmol/l)	0.62 ± 0.24	0.69 ± 0.28	0.78 ± 0.36	1.08 ± 0.95	0.0004	0.37
ApoA-I (g/l)	1.32 ± 0.18	1.36 ± 0.18	1.31 ± 0.16	1.28 ± 0.18	0.4352	0.59
ApoB (g/l)	0.68 ± 0.16	0.74 ± 0.18	0.75 ± 0.18	0.93 ± 0.37	0.0007	0.30
LDL size (Å)	272 ± 5	269 ± 5	269 ± 4	266 ± 7	0.0003	0.01

Data are means ± SD of unajusted data.

optimal and high-normal groups. Men with high BP had greater BMI, percent body fat, serum leptin, insulin, HOMA-IR and β-cell, and triglyceride and apoB than men with high-normal BP, except for serum adiponectin and LDL size, which were lower and smaller, respectively, in the high BP group than in the high-normal BP group.

Because there were substantial differences in BMI and percent body fat among the four groups, we adjusted for these two variables. After adjustment for BMI (Table 2), the difference was still significant in heart rate, serum adiponectin, and LDL size. Adjustment for percent body fat produced similar results (data not shown).

Figure 1 shows BMI-adjusted least square means of the three variables. Statistical differences among the four groups were calculated using Tukey-Kramer's procedure. As compared with men with optimal BP, those with high-normal BP had elevated heart rate, lower adiponectin concentrations, and smaller LDL size. Although there were no significant differences between the high BP and the other three groups, this may have been because of the small number of men with high BP. After adjusting for adiponectin, differences among the four groups were still significant in resting heart rate and LDL size (data not shown).

CONCLUSIONS — The major findings in the present study were that young healthy men with high-normal BP have higher heart rate, lower serum adiponectin, and smaller LDL size than those with optimal BP, even after adjustment for BMI or percent body fat. It should be noted that in the present study these observations were found in a homogeneous sample of male college students who had few confounding factors. In fact, no subject had any abnormality in electrocardiograms, a fasting glucose ≥7.0 mmol/l, hypertriglyceridemia (fasting triglyceridemia ≥4.5 mmol/l), or low HDL cholesterol (<0.9mmol/l), and no subject was currently receiving any medications. In addition, this cohort had a low prevalence of obesity (5%) and hypertension (5%) and high LDL cholesterol (6%), as previously reported (8). Finally, the proportion of current smokers was also low (5%). As a consequence, these subjects had low circulating CRP concentrations with a median value of 0.16 mg/l, which is close to a median of 0.15 mg/l in children ages 10–11 years (28).

Results from a recent prospective study (6) demonstrated that high-normal and high BP was associated with an increased risk of type 2 diabetes in 7,514 Japanese men ages 35–60 years whose BMI averaged 23.3 kg/m^2. These associations persisted even after adjustment for age and BMI. In the present study, young men with high-normal BP had lower serum adiponectin than those with normal BP after adjustment for BMI. Lower circulating levels of adiponectin have been found in individuals with obesity (10,24), type 2 diabetes (10,29), and cardiovascular disease (29,30), conditions commonly associated with insulin resistance. Hotta et al. (31) reported that circulating adiponectin concentrations decreased in parallel to the progression of insulin resistance in rhesus monkeys, which spontaneously develop obesity and frequently progress to overt type 2 diabetes. More recently, Yamauchi et al. (32) demonstrated that adiponectin administration improved insulin resistance in obese mice fed a high-fat diet and diabetic mice with lipoatrophy, suggesting that decreased levels of adiponectin are implicated in the development of insulin resistance. Taken together, it is reasonable to assume that insulin resistance may be one of reasons why high-normal BP increases the risk of type 2 diabetes (6).

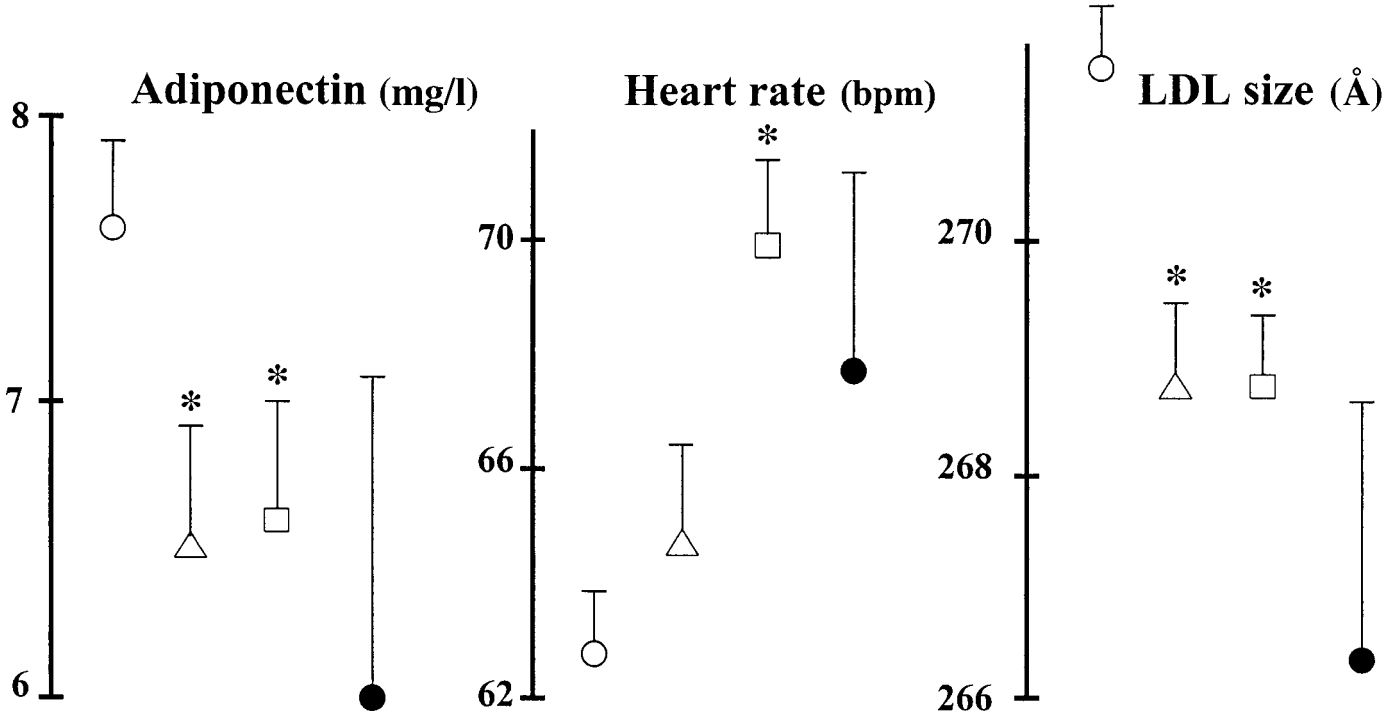

Figure 1—*BMI-adjusted least square means (±SE) of heart rate, serum adiponectin, and LDL size in young men with optimal (○; n = 83–90), normal(△; n = 45–52), high-normal (□; n = 39–46), and high (●, n = 10) BP. *P < 0.05 versus optimal BP analyzed using Tukey-Kramer's multiple comparison procedure.*

In a prospective study in elderly men and women (33), a predominance of small LDL particles was a risk factor for the future development of type 2 diabetes. This association was independent of age, sex, glucose intolerance, and BMI, but was not independent of fasting triglyceride or insulin levels. Therefore, the authors concluded that the observed association between a predominance of small LDL particle size and type 2 diabetes reflected the role of small LDL as a marker for insulin resistance rather than a causal relation between this lipoprotein phenotype and the development of diabetes. However, Haffner et al. (34) showed that decreases in LDL size are associated not only with insulin resistance, but also with a selective β-cell defect in nondiabetic subjects from the San Antonio Heart Study. In the present study, there were significant stepwise increases across the four BP status groups not only in HOMA IR, but also in HOMA β-cell concentrations, although the statistical significance disappeared after adjustment for BMI. In addition, differences among the four groups in LDL size were still significant, even after adjustment for serum adiponectin concentrations. Therefore, smaller LDL size may have reflected the role of LDL size as a marker for subtle glucose intolerance in young men in the present study, although mechanisms underlying this association are unclear.

Resting heart rate is a highly variable physiological measurement affected by many influences, including activity, stress, and stimulant substances such as nicotine and caffeine. Although we controlled for those factors as much as possible, the measurements we obtained were likely to be imprecise, which would tend to lead to an underestimation of the strength of the associations. Nonetheless, we found elevated heart rate in young men with high-normal BP as compared with those with optimal BP, even after adjusting for BMI or adiponectin. A possible explanation is that sympathetic overactivity precedes the increment in BP, overweight, and metabolic disturbances, as postulated by Julius et al. (35), although the resting heart rate is an imperfect index of sympathetic nerve activity. In addition, our results may be in keeping with observations that neurovascular dysfunction is a hallmark of the insulin-resistance syndrome and co-segregates with many of the metabolic features (36), therefore indicating that neurovascular dysfunction or sympathetic nerve dysfunction occurs early in the evolution of type 2 diabetes. Furthermore, these defects have been found in family members of type 2 diabetes and in prediabetic patients (37,38). These findings could support our notion that there may be a syndrome that long precedes the appearance of hyperglycemia or type 2 diabetes and that is characterized by elevated blood pressure within the normal range and increases in insulin resistance.

A number of studies have shown that a fast heart rate is associated with high BP and metabolic disturbance, and that it is a strong precursor of hypertension, atherosclerosis, and cardiovascular events (39). Although sympathetic overactivity seems to be responsible for these associations (40), Flanagan et al. (41) measured heart rate, insulin sensitivity (using an intravenous glucose tolerance test with minimal model), and autonomic nervous activity (using spectral analysis of heart rate variability) in young adults. They showed that insulin sensitivity is inversely associated with resting heart rate and cardiac sympathovagal balance in men, but not women. Recently, insulin sensitivity and insulin

secretion have been demonstrated to be independently related to heart rate in nondiabetic subjects in the Insulin Resistance Atherosclerosis Study (11). We have also found a significant and independent association of heart rate with HOMA IR, but not with HOMA β-cell levels (T.K., A.K., and G.Y., unpublished observations).

In conclusion, the present study demonstrates that even after adjustment for BMI, young men with high-normal BP have lower serum adiponectin concentrations, smaller LDL size, and elevated heart rate than those with optimal BP. Although we can only speculate about the mechanisms underlying these differences and recognize that they should be further investigated, we believe that young men with high-normal BP should be encouraged to modify their lifestyles to increase their insulin sensitivity and thereby avoid further development of metabolic and subsequent cardiovascular diseases.

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