Long-Term Association of Cardiovascular Risk Factors With Impaired Insulin Secretion and Insulin Resistance

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The study aim was to investigate the association of cardiovascular risk factors with insulin resistance and impaired insulin secretion in an 8-year prospective population study in nondiabetic subjects. Cardiovascular risk factors of 271 subjects aged 16 to 61 years were measured at baseline, and insulin sensitivity and acute-phase insulin secretion were assessed by an intravenous glucose tolerance test (IVGTT) and Bergman's minimal model 8 years later. In logistic regression analysis, baseline high-density lipoprotein (HDL) and very–low-density lipoprotein (VLDL) cholesterol ($P < .001$ and $P = .006$, respectively), total, low-density lipoprotein (LDL), and VLDL triglycerides ($P = .004$, $P = .048$, and $P = .002$, respectively), apolipoprotein A1 ($P = .010$), and uric acid ($P < .001$) were associated with insulin resistance after adjustment for age and the body mass index (BMI). Systolic blood pressure ($P = .042$) and VLDL cholesterol ($P = .018$) were associated with impaired insulin secretion after adjustment for age and the BMI. This 8-year longitudinal study demonstrates that dyslipidemia, high blood pressure, and uric acid are associated with insulin resistance, whereas high systolic blood pressure and VLDL cholesterol are associated with impaired first-phase insulin secretion.

INSULIN RESISTANCE and its association with metabolic abnormalities predisposing to cardiovascular disease have been a focus of intensive investigation. The importance of insulin resistance is obvious, since high fasting or post–glucose load insulin levels,[1-3] as well as insulin resistance per se,[4] have been shown to be risk factors for coronary heart disease (CHD). The insulin resistance syndrome, or syndrome X, a clustering of hyperinsulinemia, varying degrees of glucose intolerance, low high-density lipoprotein (HDL) cholesterol, high triglycerides, and hypertension, was first introduced by Reaven.[5] Recently, the concept of the insulin resistance syndrome has been extended to include also abnormalities of the fibrinolytic system, an elevation of uric acid, and the presence of small, dense low-density lipoprotein (LDL) particles.[6] In addition to insulin resistance, impaired insulin secretion is known to be an important underlying defect in the development of type 2 diabetes mellitus.[7]

Cross-sectional and longitudinal studies have demonstrated an association of hyperinsulinemia, as well as directly measured insulin resistance, with lipoprotein abnormalities, ie, low HDL cholesterol, high total or very–low-density lipoprotein (VLDL) triglycerides, and small, dense LDL.[8-15] Similarly, hypertension has been shown to be an insulin-resistant state,[16] but the association of insulin sensitivity with blood pressure in normotensive subjects has been inconsistent.[17,18] In contrast, information on the association of impaired insulin secretion and cardiovascular risk factors is limited and contradictory.[19,20] Two studies suggest that impaired insulin secretion is associated with visceral adiposity and weight gain.[21,22] In addition, our knowledge about the association of cardiovascular risk factors with insulin resistance is still limited, and to our knowledge, longitudinal data on cardiovascular risk factors predicting impaired insulin secretion are not available. Although a longitudinal design cannot prove causality, useful information can be obtained on studies concerning the temporal relation of insulin resistance and/or impaired insulin secretion with cardiovascular risk factors. Therefore, we measured cardiovascular risk factors at baseline in 271 nondiabetic subjects and correlated them with insulin sensitivity and insulin secretion determined 8 years later.

SUBJECTS AND METHODS

Study Population and Protocol

The subjects for the study were participants in our previous population study to investigate cardiovascular morbidity and its risk factors in diabetic and nondiabetic families with and without CHD.[23] Altogether, 745 subjects participated in the baseline study in 1983 to 1985 and 615 subjects (82.5%) in the follow-up study in 1992 to 1993. Altogether, 271 participants of the follow-up study volunteered for an intravenous glucose tolerance test (IVGTT).

The baseline and follow-up study included the determination of serum lipids and lipoproteins and uric acid, an oral glucose tolerance test (OGTT), measurement of blood pressure, weight, and height, as well as an interview on the subject's history of smoking, physical activity, and alcohol intake. Fasting samples (12-hour fast) were taken for the determination of lipids, lipoproteins, uric acid, plasma glucose, and insulin, and subsequently, all subjects underwent an OGTT (75 g glucose). Blood samples for the determination of plasma glucose and insulin were obtained during fasting and 1 and 2 hours after the glucose load. Classification of the glucose tolerance status was performed according to World Health Organization criteria.[24] If the fasting plasma glucose value was less than 7.8 mmol/L and the 2-hour value was greater than 7.8 and maximally 11.0 mmol/L, the subject was classified as having impaired glucose tolerance. Blood pressure was measured in the sitting position after a 5-minute rest, with a mercury sphygmomanometer. Both systolic and diastolic blood pressure were read to the nearest 2 mm Hg. Systolic blood pressure was determined from the appearance of the Korotkoff sound (phase I), and diastolic blood pressure from the disappearance of the Korotkoff sound (phase V). The measurement was performed twice 1.5 minutes apart, and the second value was used in statistical analyses. A subject was classified as hypertensive if the systolic blood pressure was 160 mm Hg or higher or diastolic blood pressure was 95 mm Hg or higher or if the patient was

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Submitted March 27, 1999; accepted April 13, 2000.

Supported by grants from the Finnish Heart Research Foundation, Aarne and Aili Turunen Foundation, Aarne Koskelo Foundation, Orion Research Foundation, and Academy of Finland.

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0026-0495/00/4910-0010$10.00/0

doi:10.1053/meta.2000.9514

receiving medication for hypertension. Weight and standing height were measured without shoes in light clothes. The relative weight was expressed as the body mass index (BMI), calculated as weight in kilograms divided by height in meters squared. Smoking was defined as current smoking. Alcohol intake was determined as a dichotomy of consumers or nonconsumers. Subjects were classified as physically active if their work was heavy (for example, lumberjacking or physically heavy industrial work) or if they were physically active in leisure time (at least 30 minutes of activity at least 3 times per week). In the follow-up study, cardiovascular risk factors were determined according to the same protocol already described and an IVGTT was performed 2 to 12 weeks (average, 4 weeks) later at the second visit.

IVGTT

In the follow-up study, the subjects underwent an IVGTT using the frequently sampled IVGTT protocol. Between 8 and 9 AM after a 12-hour fast, an indwelling cannula was inserted into an antecubital vein for injection of glucose and insulin. Another cannula for blood sampling was inserted into the antecubital vein of the opposite arm. Two successive fasting blood samples (5 minutes apart) were obtained for measurement of blood glucose and plasma insulin. An intravenous glucose bolus (0.3 g glucose/kg body weight as a 50% solution administered over 2 minutes) was then injected via the cannula in the arm opposite the sampling arm. Additional samples for blood glucose and plasma insulin measurements were taken at 4, 6, 8, 10, 19, 22, 29, 37, 67, 90, and 180 minutes. At 20 minutes, an intravenous injection of regular insulin (0.03 U/kg) was administered to increase the accuracy of the modeling analyses.[25,26] Glucose utilization was analyzed using the Minimal Model of glucose disappearance of Bergman et al.[27] The equations of this model provide measures of the sensitivity of glucose elimination to insulin (insulin sensitivity index, S_I, inversely proportional to insulin resistance). Estimates of S_I from this model have been validated against the glucose clamp technique.[28]

Analytic Methods

At the baseline study, plasma glucose was determined by glucose dehydrogenase (Merck Diagnostica, Darmstadt, Germany). For the determination of plasma insulin, blood samples were obtained in chilled tubes and the plasma was separated immediately. Samples were stored at −20°C and analyzed by radioimmunoassay (Phadeseph; Pharmacia Diagnostics, Uppsala, Sweden). The VLDL fraction was separated by ultracentrifugation. Serum HDL cholesterol was determined after precipitation of VLDL and LDL fractions with dextran sulfate and $MgCl_2$. The concentration of LDL cholesterol was calculated as the difference between the bottom fractions. HDL_2 and HDL_3 subfractions were separated by ultracentrifugation, and the top and bottom fractions were separated by a tube-slicing technique. A commercial enzymatic method was used in the determination of cholesterol (Monotest; Boehringer, Mannheim, Germany) and triglycerides (Test-Combination Triglyceride; Boehringer). Commercial control sera were used to standardize the measurements of cholesterol and triglycerides (Seronorm, Seronorm Lipids; Nycomed, Oslo, Norway). Serum apolipoproteins A1 and B were determined from serum samples stored at −70°C. The determinations were performed by a commercial immunochemical method based on the measurement of immunoprecipitation at a wavelength of 340 nm (Kone Instruments Division, Kone, Espoo, Finland). Serum uric acid was determined by the uricase method (Boehringer) in association with a colorimetric reaction (Hitachi 705 automatic analyzer; Hitachi, Tokyo, Japan).

Statistical Methods

Statistical analyses were performed with SPSS-PC+ programs (SPSS, Chicago, IL). Age- and BMI-adjusted levels of baseline cardiovascular risk factors in tertiles of S_I were calculated with analysis of covariance (ANCOVA), and when the groups differed significantly ($P < .05$), other tertiles were compared with the lowest tertile. Insulin secretion was determined as the insulin area under the first 10 minutes of the insulin concentration curve during the IVGTT. Logistic regression analysis was used to analyze cardiovascular risk factors associated with S_I and the area under the insulin curve (AUC) during the first 10 minutes of the IVGTT. S_I was dichotomized using the lowest tertile as a cut off point for insulin resistance ($S_I \leq 2.85 \times 10^{-4}$/min/mU/mL; S_I range, 0 to 12.10×10^{-4}/min/μU/mL). Similarly, the AUC was dichotomized using the lowest tertile (AUC ≤ 1,584 min · pmol/L; AUC range, 280 to 8,662 min · pmol/L) as a cutoff point for impaired first-phase insulin secretion. The skewed distribution of triglycerides and insulin was corrected with logarithmic transformation.

The study was approved by the Ethics Committee of Kuopio University Hospital.

RESULTS

Baseline characteristics of the subjects (N = 271) are shown in Table 1. At baseline, the mean age of the participants was 33.5 years (range, 16 to 61) and the mean BMI was 24.0 kg/m^2 (range, 17.1 to 42.6). Twenty-seven (10.0%) of the subjects were hypertensive and 6 (2.2% of them had impaired glucose tolerance at baseline. In the follow-up study, the mean S_I was $4.26 \pm 0.15 \times 10^{-4}$/min/μU/mL (range, 0 to 12.10) and the mean AUC was 2,435 ± 125 min · pmol/L (range, 280 to 8,662).

Figure 1 illustrates age- and BMI-adjusted baseline cardiovascular risk factors in tertiles of S_I measured at the follow-up study. Compared with the subjects in the middle and highest

Table 1. Baseline Characteristics of the Subjects (N = 271)

Characteristic	Mean ± SEM (range)
Age (yr)	33.5 ± 0.7 (16-61)
BMI (kg/m^2)	24.0 ± 0.2 (17.1-42.6)
Smokers (%)	26.6
Physically active (%)	51.3
Alcohol users (%)	58.7
Impaired glucose tolerance (%)	2.2
Hypertension (%)	10.0
Systolic BP (mm Hg)	129.7 ± 1.0
Diastolic BP (mm Hg)	77.9 ± 0.7
Cholesterol (mmol/L)	
Total	5.94 ± 0.08
HDL	1.38 ± 0.02
HDL_2	1.01 ± 0.02
HDL_3	0.37 ± 0.01
LDL	3.82 ± 0.06
VLDL	0.75 ± 0.02
Triglycerides (mmol/L)	
Total	1.14 ± 0.04
HDL	0.06 ± 0.01
LDL	0.28 ± 0.01
VLDL	0.80 ± 0.03
Apolipoprotein A1 (g/L)	1.42 ± 0.01
Apolipoprotein B (g/L)	1.10 ± 0.03
Plasma glucose (mmol/L)	
Fasting	5.0 ± 0.1
2-hour	5.2 ± 0.1
Insulin (pmol/L)	
Fasting	72.4 ± 2.1
2-hour	267.1 ± 12.5
Uric acid (μmol/L)	275.5 ± 4.2

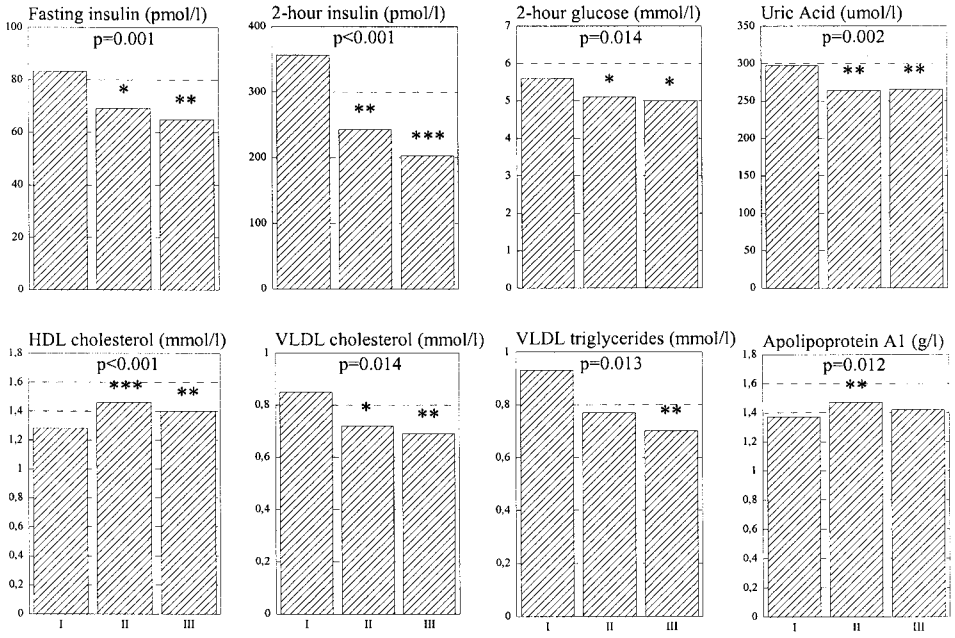

Fig 1. Age- and BMI-adjusted baseline cardiovascular risk factors in tertiles of S_I (I, II, and III) compared with the lowest tertile of S_I (I).

tertiles of S_I, subjects in the lowest tertile had significantly higher fasting and 2-hour insulin, 2-hour glucose, uric acid, and VLDL cholesterol and significantly lower HDL cholesterol. VLDL triglycerides were higher in the lowest tertile of S_I versus the highest tertile. Apolipoprotein A1 was higher in the middle tertile of S_I, but did not differ in the highest tertile compared with the lowest tertile of S_I.

Table 2 shows the association of baseline cardiovascular risk factors adjusted for age and BMI with insulin resistance ($S_I \leq 2.85 \times 10^{-4}$/min/μU/mL) or impaired insulin secretion

Table 2. Baseline Cardiovascular Risk Factors Adjusted for Age and BMI Predicting Insulin Resistance (lowest tertile of S_I, $\leq 2.85 \times 10^{-4}$/min/μU/mL) or Impaired Insulin Secretion (lowest tertile of AUC in IVGTT, ≤1,584 min · pmol/L) by Logistic Regression Analyses

	Insulin Resistance			Impaired Insulin Secretion		
Risk Factor	OR	95% CI	*P*	OR	95% CI	*P*
Smoking	0.93	0.48-1.79	NS	1.23	0.69-2.18	NS
Physical activity	0.55	0.31-0.96	.037	1.28	0.77-2.14	NS
Alcohol	1.17	0.66-2.06	NS	1.59	0.94-2.69	NS
Systolic BP	1.00	0.98-1.01	NS	1.02	1.00-1.04	.042
Diastolic BP	1.03	0.99-1.06	.072	1.01	0.98-1.04	NS
Cholesterol						
Total	0.97	0.77-1.23	NS	0.99	0.79-1.23	NS
HDL	0.13	0.04-0.39	<.001	1.12	0.48-2.61	NS
HDL_2	0.13	0.04-0.39	<.001	1.12	0.49-2.57	NS
HDL_3	1.54	0.07-33.29	NS	0.96	0.05-17.45	NS
LDL	0.99	0.75-1.30	NS	1.10	0.84-1.42	NS
VLDL	2.97	1.35-6.51	.006	0.39	0.18-0.85	.018
Triglyceride*						
Total	2.47	1.34-4.58	.004	0.91	0.52-1.60	NS
HDL	0.91	0.66-1.25	NS	1.00	0.73-1.37	NS
LDL	2.08	1.01-4.30	.048	0.76	0.39-1.49	NS
VLDL	2.17	1.31-3.58	.002	0.93	0.60-1.44	NS
Apolipoprotein A1	0.19	0.05-0.68	.010	1.03	0.35-3.01	NS
Apolipoprotein B	1.55	0.81-2.97	NS	0.87	0.45-1.65	NS
Plasma glucose						
Fasting	1.53	0.95-2.45	NS	1.63	1.03-2.57	.035
2-hour	1.37	1.09-1.70	.007	1.13	0.93-1.36	NS
Insulin						
Fasting*	3.67	1.76-7.65	.002	0.29	0.14-0.58	<.001
2-hour*	2.97	1.85-4.75	<.001	0.62	0.42-0.92	.018
Uric acid	1.01	1.00-1.02	<.001	1.00	0.99-1.01	NS
Weight change	1.09	1.04-1.15	<.001	0.94	0.89-0.98	.004

*Logarithmic transformation.

(AUC ≤ 1,584 min · pmol/L) in the follow-up study analyzed by logistic regression analysis. In logistic regression, physical activity, HDL and HDL_2 cholesterol, VLDL cholesterol, total, LDL, and VLDL triglycerides, apolipoprotein A1, 2-hour glucose, fasting and 2-hour insulin, and uric acid were associated with insulin resistance. Weight change (difference between follow-up and baseline weight) had a significant positive association with insulin resistance. Systolic blood pressure, VLDL cholesterol, fasting glucose, and fasting and 2-hour insulin had a significant association with impaired insulin secretion. Weight change had a significant negative association with impaired insulin secretion.

To determine if baseline cardiovascular risk factors were associated with insulin resistance 8 years later also in the more insulin-sensitive subgroup, univariate logistic regression analyses were performed by excluding subjects in the highest tertile of baseline fasting insulin (≥77.0 pmol/L; the most insulin-resistant subjects). After adjustment for age and BMI, total and VLDL triglycerides (odds ratio in total and VLDL triglycerides [OR] = 2.78, 95% confidence interval [CI] = 1.24, 6.26, $P = .013$ and OR = 2.18, 95% CI = 1.14-4.18, $P = .018$, respectively), 2-hour plasma glucose (OR = 1.61, 95% CI = 1.18-2.20, $P = .002$), and fasting and 2-hour insulin (OR = 8.24, 95% CI = 1.79-37.90, $P = .002$ and OR = 4.79, 95% CI = 2.29-9.95, $P < .001$, respectively) were still positively associated with insulin resistance, and HDL_2 cholesterol (OR = 0.23, 95% CI = 0.05-0.94, $P = .041$) and physical activity (OR = 0.45, 95% CI = 0.22-0.92, $P = .028$) had a significant inverse association with insulin resistance. In contrast, HDL and VLDL cholesterol, LDL triglycerides, apolipoprotein A1, and uric acid were not significantly related to insulin resistance.

Furthermore, the associations of cardiovascular risk factors measured at follow-up with insulin resistance were analyzed. After adjustment for age and BMI, diastolic blood pressure (OR = 1.03, 95% CI = 1.00-1.06, $P = .056$), HDL (OR = 0.05, 95% CI = 0.02-0.18, $P < .001$) and VLDL cholesterol (OR = 4.36, 95% CI = 2.05-9.27, $P < .001$), total (OR = 5.40, 95% CI = 2.65-11.01, $P < .001$), HDL (OR = 8.21, 95% CI = 2.90-23.25, $P < .001$), LDL (OR = 3.55, 95% CI = 1.54-8.13, $P = .001$), and VLDL triglycerides (OR = 3.01, 95% CI = 1.83-4.95, $P < .001$), apolipoprotein A1 (OR = 0.05, 95% CI = 0.01-0.27, $P < .001$), apolipoprotein B (OR = 4.92, 95% CI = 1.66-14.53, $P = .004$), fasting and 2-hour glucose (OR = 2.48, 95% CI = 1.42-4.31, $P = .001$ and OR = 1.43, 95% CI = 1.16-1.76, $P < .001$, respectively), and fasting and 2-hour insulin (OR = 12.92, 95% CI = 5.13-32.54, $P < .001$ and OR = 5.06, 95% CI = 2.79-9.20, $P < .001$, respectively) measured at follow-up study were significantly associated with insulin resistance in logistic regression analyses.

Table 3 shows baseline cardiovascular risk factors associated with the simultaneous presence of insulin resistance and impaired insulin secretion (the lowest tertiles of both S_I and AUC in IVGTT, n = 13) by logistic regression analysis. In univariate analyses, the age, BMI, alcohol intake, diastolic blood pressure, total, LDL, and VLDL triglycerides, fasting and 2-hour glucose, 2-hour insulin, and uric acid were significantly associated with the simultaneous presence of insulin resistance and impaired insulin secretion. After adjustment for age and BMI, the associations with alcohol, total and VLDL triglycerides, and fasting and 2-hour glucose remained significant.

Table 4 shows the results of multiple logistic regression analysis. The independence of the association of insulin resistance with either HDL cholesterol or VLDL triglycerides was analyzed by including in each model an additional cardiovascular risk factor. HDL cholesterol was significantly associated with insulin resistance when adjustment was made consequently for age, BMI, fasting insulin, diastolic blood pressure, uric acid, and VLDL cholesterol (models 1a to 4a). The association of VLDL triglycerides with insulin resistance was significant after adjustment for age, BMI, and fasting insulin (models 1b and 2b), but after including diastolic blood pressure, uric acid, and VLDL cholesterol in the model, the association was no longer significant (models 3b and 4b). Diastolic blood pressure was not associated with insulin resistance in either model 2a or 2b. Uric acid was significantly associated with insulin resistance after adjustment for age, BMI, fasting insulin, and VLDL triglycerides ($P = .024$; model 3b). Multiple logistic regression analysis was also performed to analyze the independence of the association of impaired insulin secretion (lowest tertile of AUC in IVGTT) with cardiovascular risk factors by including in each model an additional variable (data not shown). After adjustment for age, BMI, and fasting insulin, VLDL cholesterol no longer had an association with impaired insulin secretion ($P = .078$). In the model including age, BMI, fasting insulin, VLDL cholesterol, and systolic blood pressure, systolic blood pressure still had a significant association with impaired insulin secretion ($P = .018$), whereas VLDL cholesterol had no association ($P = .083$). In the model including age, BMI, fasting insulin, VLDL cholesterol, systolic blood pressure, and fasting glucose, the association of systolic blood pressure ($P = .026$) and fasting glucose ($P = .004$) with impaired insulin secretion remained significant.

DISCUSSION

Our population-based follow-up study demonstrates for the first time that high systolic blood pressure, but not dyslipidemia, is associated with impaired first-phase insulin secretion in nondiabetic middle-aged subjects. The association of dyslipidemia (low HDL and HDL_2 cholesterol and high total, LDL, and VLDL triglycerides), high blood pressure, and high uric acid with insulin resistance was consistent during the 8-year follow-up study. These associations with insulin resistance remained essentially unchanged when the most hyperinsulinemic subjects at baseline were excluded.

Limited and inconsistent data are available on the association of cardiovascular risk factors with impaired insulin secretion.[19,20] We demonstrated that high blood pressure was associated independently of other cardiovascular risk factors with impaired insulin secretion evaluated by the insulin AUC during the first 10 minutes of the IVGTT 8 years later. Sinagra et al[20] demonstrated that blood pressure was not related to insulin secretion, defined as fasting or glucose-stimulated C-peptide levels, in 21 female subjects. In contrast, Istfan et al[29] reported that systolic blood pressure was positively correlated with insulin secretion assessed as meal-stimulated C-peptide excretion in 72 middle-aged subjects. In the previous studies, the stimulated C-peptide level or urinary excretion were used,

Table 3. Cardiovascular Risk Factors Associated With the Simultaneous Occurrence of Insulin Resistance (lowest tertile of S_I, ≤2.85 × 10^{-4}/min/μU/mL) and Impaired Insulin Secretion (lowest tertile of AUC in IVGTT, ≤1,584 min · pmol/L) by Logistic Regression Analyses

	Univariate			Adjusted for Age and BMI		
Risk Factor	OR	95% CI	*P*	OR	95% CI	*P*
Age	1.05	1.01-1.10	.048			
BMI	1.30	1.06-1.51	.004			
Sex	0.58	0.18-1.85	NS	0.45	0.12-1.74	NS
Smoking	1.15	0.33-4.03	NS	1.07	0.25-4.51	NS
Physical activity	0.73	0.23-2.32	NS	0.81	0.22-2.98	NS
Alcohol	4.90	1.03-23.20	.045	8.72	1.17-65.17	.035
Systolic BP	1.03	0.99-1.06	.067	1.01	0.95-1.04	NS
Diastolic BP	1.06	1.01-1.11	.024	1.02	0.96-1.09	NS
Cholesterol						
Total	1.28	0.83-1.98	NS	1.02	0.58-1.78	NS
HDL	0.29	0.04-2.38	NS	0.26	0.02-3.17	NS
HDL_2	0.25	0.03-2.27	NS	0.27	0.02-3.31	NS
HDL_3	2.44	0.01-941.14	NS	0.78	0.01-1,027.58	NS
LDL	1.52	0.88-2.63	NS	1.14	0.58-2.25	NS
VLDL	1.50	0.32-7.14	NS	1.00	0.61-1.64	NS
Triglyceride*						
Total	6.11	1.75-21.37	.005	4.30	1.08-17.13	.038
HDL	1.69	0.85-3.45	NS	1.24	0.59-2.62	NS
LDL	3.97	1.01-16.67	.049	2.59	0.62-10.75	NS
VLDL	5.18	1.72-15.59	.003	4.16	1.21-14.25	.023
Apolipoprotein A1	1.47	0.14-15.58	NS	3.53	0.19-65.04	NS
Apolipoprotein B	1.90	0.61-5.88	NS	1.57	0.35-6.94	NS
Plasma glucose						
Fasting	5.32	2.04-14.29	<.001	4.11	1.48-11.39	.007
2-hour	3.03	1.69-5.55	<.001	3.05	1.51-6.18	.002
Insulin						
Fasting*	1.75	0.42-7.14	NS	0.76	0.13-4.30	NS
2-hour*	2.77	1.06-7.25	.037	2.05	0.73-5.72	NS
Uric acid	1.01	1.00-1.02	.041	1.01	0.99-1.02	NS

*Logarithmic transformation.

which are indirect measures of insulin secretion and do not distinguish first- and second-phase insulin secretion. In the present study, we evaluated first-phase insulin secretion by an IVGTT. Moreover, the number of study subjects in previous studies was smaller than in the present study. Our findings support the hypothesis that disturbances in insulin secretion and blood pressure regulation are related, although causality cannot be proven. However, it is obvious that the associations of insulin

Table 4. Baseline Cardiovascular Risk Factors Associated With Insulin Resistance (S_I ≤2.85 × 10^{-4}/min/μU/mL) by Multivariate Logistic Regression Analyses

	Model 1a		Model 2a		Model 3a		Model 4a	
Risk Factor	B	*P*	B	*P*	B	*P*	B	*P*
Age	0.037	.015	0.030	NS	0.034	.043	0.033	.048
BMI	0.160	.002	0.142	.006	0.126	.015	0.125	.0175
HDL cholesterol	−1.819	.001	−1.796	.001	−1.546	.007	−1.466	.014
Fasting insulin*	1.196	.002	1.176	.003	1.172	.004	1.146	.005
Diastolic blood pressure	—	—	0.020	NS	0.014	NS	0.014	NS
Uric acid	—	—	—	—	0.005	.059	0.004	.073
VLDL cholesterol	—	—	—	—	—	—	0.200	NS
	Model 1b		Model 2b		Model 3b		Model 4b	
Age	0.017	NS	0.010	NS	0.016	NS	0.016	NS
BMI	0.180	<.001	0.164	.002	0.144	.006	0.141	.007
VLDL triglycerides*	0.607	.022	0.559	.037	0.373	NS	0.237	NS
Fasting insulin*	1.105	.004	1.101	.005	1.113	.005	1.103	.005
Diastolic blood pressure	—	—	0.018	NS	0.013	NS	0.013	NS
Uric acid	—	—	—	—	0.005	.024	0.005	.028
VLDL cholesterol	—	—	—	—	—	—	0.332	NS

*Logarithmic transformation.

secretion and insulin resistance with blood pressure are complex.

We did not find any significant associations of lipoproteins with impaired first-phase insulin secretion. To our knowledge, there are no previous data on this issue. Furthermore, we did not find associations of smoking, alcohol, or physical activity with impaired insulin secretion, suggesting that these factors are not important determinants of insulin secretion. A partly similar finding was reported recently by Clausen et al[19] in young healthy subjects. In their study, only 10% of the variation in the acute insulin response measured by an IVGTT was explained by anthropometric and environmental determinants (alcohol, smoking, saturated fat intake, and use of oral contraceptives) in 380 young subjects. Finally, in our study, weight loss during the 8-year follow-up study was associated with impaired first-phase insulin secretion. Previous studies have reported opposite results in different ethnic groups, demonstrating that impaired insulin secretion leads to weight gain and has adverse effects on body fat distribution.[21,22] In the prospective study of 97 normoglycemic Pima Indians, insulin secretion assessed by either an OGTT or IVGTT was inversely associated with the rate of weight gain over more than 3 years, indicating that impaired insulin secretion was a marker of an increased risk for weight gain.[21] However, in that study, only 10% to 12% of the variance in weight gain was attributable to differences in insulin secretion. In the 5-year follow-up study of Japanese-American men, reduced insulin secretion preceded visceral fat accumulation in 137 nondiabetic subjects.[22] These data suggest that insulin's role to regulate weight and fat distribution differs among ethnic groups.

Previous cross-sectional studies have demonstrated an association of hyperinsulinemia with low HDL cholesterol and high total and VLDL triglycerides.[30-32] Insulin resistance per se, determined by the euglycemic clamp technique or Minimal Model method, is related to low HDL cholesterol and high total and VLDL triglycerides in diabetics[17,33] and nondiabetic subjects.[9,11,12,33-36] Essential hypertension has been demonstrated to be an insulin-resistant state in non-obese nondiabetic[16,37] and diabetic[38] subjects in cross-sectional studies. However, data concerning the relationship between blood pressure and insulin resistance in normotensive subjects have been inconsistent. In the San Antonio Heart Study, fasting insulin levels were correlated cross-sectionally with blood pressure.[39] In the study by Blonck et al,[17] insulin sensitivity measured by the clamp technique was not related to blood pressure in 46 normotensive non–insulin-dependent diabetics. Also, Arslanian and Suprasongsin[36] reported that in 20 healthy prepubertal children diastolic blood pressure was negatively correlated with insulin sensitivity. In contrast, in the study by Godsland et al[9] including 158 healthy men, insulin resistance measured by the Minimal Model method was associated with blood pressure and this association was dependent on age and BMI, similar to the present study. In general, the association of blood pressure and insulin resistance has been weaker than the association of insulin resistance and dyslipidemia.[9]

Hyperuricemia has been suggested to be independently associated with insulin resistance in cross-sectional studies.[40,41] Facchini et al[40] demonstrated that urinary uric acid clearance decreased in proportion to an increase in insulin resistance, leading to an increase in serum uric acid, indicating that insulin resistance would have a major role in uric acid clearance at the level of the kidney. On the other hand, Vuorinen-Markkola and Yki-Järvinen[41] have suggested that increased lipolysis and VLDL production lead to an overproduction of uric acid.

What are the explanations for the association of cardiovascular risk factors, particularly dyslipidemia and hyperuricemia, with insulin resistance in our prospective population-based study? The possibility that cardiovascular risk factors actually cause insulin resistance has some support in in vitro studies, which have demonstrated that VLDL or triglycerides may impair insulin receptor function or inhibit glucose transport.[42,43] Although this could be interpreted to indicate that insulin resistance is a consequence of lipoprotein abnormalities, no previous population-based studies are available to substantiate this notion. In fact, prospective studies have suggested an opposite association.[13,14] Findings from the San Antonio Heart Study demonstrated that high fasting plasma insulin preceded the development of hypertriglyceridemia and low HDL cholesterol, as well as high blood pressure, 8 years later.[13] Our previous prospective study of elderly nondiabetic subjects has shown that baseline hyperinsulinemia is a predictor of hypertriglyceridemia, high apolipoprotein B, low apolipoprotein A1, a low LDL cholesterol to apolipoprotein B ratio, and a low HDL cholesterol to apolipoprotein A1 ratio 3.5 years later.[14] In the Honolulu Heart Program including 3,562 elderly Japanese-American men, triglycerides, BMI, and hypertension both cross-sectionally and 25 years earlier were associated independently with hyperinsulinemia.[15]

In the present study, we did not measure insulin resistance at baseline. Therefore, our findings do not exclude the possibility that adverse changes in cardiovascular risk factors could precede the development of insulin resistance, because our results remained practically unchanged after exclusion of the most hyperinsulinemic (the most insulin-resistant) subjects from statistical analyses of the data. However, it is more probable that the subjects with low insulin sensitivity in the follow-up study were insulin-resistant already 8 years earlier, because they had hyperinsulinemia already at baseline (Fig 1). However, in an epidemiologic study setting, it is difficult to draw definite conclusions on the cause-and-effect relationships. Our results confirm the data of the Honolulu Heart Program[15] that insulin resistance is persistently associated with adverse metabolic changes. Moreover, our results indicate that these unfavorable metabolic manifestations are present already at the age of 30 years, similar to the study by Arslanian and Suprasongsin,[36] who demonstrated these associations already in childhood.

No previous studies are available correlating cardiovascular risk factors simultaneously with both insulin resistance and impaired insulin secretion. In the present study, total and VLDL triglycerides, alcohol intake, and fasting and 2-hour glucose were associated with the simultaneous presence of impaired insulin secretion and insulin resistance after adjustment for age and BMI. Because these variables were not associated with impaired insulin secretion alone, the association was probably due to their association with insulin resistance. The simulta-

neous occurrence of impaired insulin secretion and insulin resistance is a marker of the prediabetic state[7] in these subjects. The strong association of fasting and 2-hour glucose levels with the impairment of insulin action and insulin secretion is in agreement with this notion, indicating that these subjects are at increased risk to develop diabetes.

We have used the lowest tertile of insulin sensitivity and first-phase insulin secretion to classify subjects with insulin resistance or impaired insulin secretion. However, the cutoff points for the lowest tertile of insulin sensitivity and insulin secretion do not necessarily classify these subjects reliably as those with insulin resistance or impaired insulin secretion. We had to apply these cutoff points because there are no data on the distribution of the insulin sensitivity index or insulin secretion measured by the Minimal Model in the Finnish population of the same age and gender distribution.

In summary, our results demonstrate that dyslipidemia, elevated levels of glucose and insulin, and hyperuricemia are associated with insulin resistance both cross-sectionally and 8 years earlier in nondiabetic middle-aged subjects. In contrast, only high systolic blood pressure is associated with impaired first-phase insulin secretion.

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