Table 3
Stepwise multiple regression analysis of factors predicting total hospital charges in 316 patients with acute ischemic stroke

Factors	Unit	β	SE	*P*	Partial R^2
LOHS	Days	0.0223	0.00102	0.0001	0.5993
Surgery	Yes/no	0.4906	0.08221	0.0001	0.0482
Cerebral angiography	Yes/no	0.0930	0.04379	0.03	0.0050

Variables not retained in the model are age, severity on admission, diabetes, and outcome.

Medicare Prospective Payment System in the United States provides a strong incentive for hospitals to discharge patients as quickly as possible. This payment system reimburses hospitals a fixed amount of money for a designated diagnosis-related group, regardless of the actual resources used. This system was originally established for Medicare patients, and has spread into many other insurance schemes. On the other hand, hospitals in Japan are reimbursed through the universal insurance scheme on a fee-for-service basis, with all prices fixed by a nationally standardized, itemized, and minutely defined schedule. Prior to the time of this study, however, there was little economic incentive for hospitals to reduce LOHS [29]. Because of the low co-payment for individual patients and the absence of a discharge policy, long-term care services in Japan were usually offered by the same hospitals that provided the acute care services. Thus, it is possible that patients tended to stay in hospitals longer than necessary. In 1995, the Japanese Ministry of Health, Labor, and Welfare started to monitor LOHS in major hospitals and encourage doctors to shorten it. This study was performed just after this monitoring system was introduced in Japan, and its findings indicated that the LOHS of stroke patients is still longer in Japan than in other developed countries. Japanese doctors now seem to be searching for the optimal method of shortening LOHS without decreasing the quality of medical service. Further time and effort may be necessary to establish this new reimbursement system.

We found that charges rose with surgery, and performance of angiography was also an independent contributor to increased charge. These factors may represent severity of the illness. An increase in the number of angiographies performed was related to an increase in LOHS (data not shown), suggesting the presence of complicated disease conditions, such as advanced cerebral atherosclerosis or multiple embolism, in these patients. Patients with severe stroke tend to require more medication and a longer hospital stay than those with stroke of lesser severity. As a result, the medical charges for severely ill patients may go higher. In our subjects, the charge increased with elevating stroke severity in the univariate analysis (Table 2), but multiple regression analysis did not reveal that stroke severity was an independent determinant of hospital charge (Table 3). This can be attributed to the increase in fatality in acute phase among patients with the most severe strokes, which would result in a lower mean charge. Thus, although LOHS was not a direct marker of stroke severity, care must be taken when shortening LOHS to avoid an increase in the risk to patients.

There are several potential limitations to the findings in our study. First, this single-center study was conducted in a large emergency-based setting that serves as tertiary referral care center for the metropolitan area of Fukuoka. Thus, generalization of our findings to other patient populations may not be appropriate. However, the inpatient treatment and charges at this hospital were similar to those of general hospitals in Japan; the average total charge per stroke inpatient in our country was approximately 1 million yen ($8333) in 1998 [1], which was similar to that in the present study. This suggests that the findings of our study represent the actual circumstances of inpatient charges for ischemic stroke in Japan. Second, we retrospectively ascertained inpatient charges without information relating charges to costs. Charges

do not necessarily reflect costs incurred in the production of hospital care. Frinkler et al. [39], who investigated inpatient charges in the United States, found that charges did not necessarily reflect the real costs incurred, because hospitals often shifted charges from uninsured patients to insured patients in order to maximize revenues. In Japan, however, the price of each service is subject to the standard national fee schedule. In regard to medical care, all residents in Japan are covered by one of the health insurance programs; they must join the program that is offered by their local governments or trade associations. Cost-sharing requirements vary with the program (e.g. 10% for employees and 30% for others), but the limit of cost sharing for all programs was 54 000 yen ($450) per month until 1996, and 60 000 yen ($500) from 1997 until the time of the study. The health insurance covers almost all medical treatment and fees for medical providers. Hospitals are not allowed to raise prices or to do balanced billing. Thus, it is certain that the charge for each service category reflected the price of the service in our study. Third, this study had a retrospective design, without a proper control group. This suggests that our findings were likely conservative. Nonetheless, we believe that our findings should help to clarify the factors contributing to hospital charges for stroke patients. Large prospective studies employing recent stroke patients will be needed to further clarify the relationship between these two phenomena.

In conclusion, the findings of our study suggest that LOHS is the key contributory predictor of hospital charges for ischemic stroke patients in Japan [25,29]. This study may be of value to other developed countries, and some developing countries, that are trying to provide comprehensive health services to a large, aging population, while at the same time, trying to avoid overburdening the state with enormous welfare bills that retard economic development. The apparent implication for policy makers is that reductions in the charges incurred for treatment of stroke are most likely to be achieved by shortening LOHS, and LOHS is most likely to be shortened by altering the reimbursement policies.

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Risk factors for renal glomerular and vascular changes in an autopsy-based population survey: The Hisayama Study

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Risk factors for renal glomerular and vascular changes in an autopsy-based population survey: The Hisayama Study.

Background. Information of the effect of cardiovascular risk factors on renal glomerular and vascular changes is scarce in the general population.

Method. Between 1962 and 1994, 1394 autopsies were performed in Hisayama, for a total autopsy rate of 80%. Of these, 839 individuals who preserved adequate renal tissues and had recent health examinations data before death were eligible for the present study. We examined the degree of glomerular sclerosis, renal arteriolar hyalinosis, and arteriosclerosis, and evaluated their risk factors by means of a logistic regression model.

Results. The development of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis were 16%, 16%, and 18% in men, respectively, and 27%, 15%, and 24% in women, respectively. All these frequencies increased linearly with advancing age. In the multivariate analysis, both age and systolic blood pressure were significant independent risk factors for almost all these glomerular and vascular changes. In addition, glucose intolerance and proteinuria for men were found to be significant risk factors for glomerular sclerosis. Elevated total cholesterol levels significantly increased the risk of arteriolar hyalinosis in men. Electrocardiogram (ECG) abnormalities were an independent risk factor for arteriosclerosis in both men and women, and proteinuria was an additional risk factor in women. Alcohol intake tended to have a protective effect on glomerular sclerosis and arteriosclerosis in women.

Conclusion. Our data confirmed that age and systolic blood pressure are common risk factors for all glomerular and renal vascular changes in the general population. In addition, glucose intolerance, total cholesterol, ECG abnormalities, and proteinuria affect either glomerular or vascular changes.

Key words: glomerular sclerosis, arteriolosclerosis, arteriosclerosis, diabetic nephropathy, nephrosclerosis, risk factor, population-based study, autopsy.

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Despite recent advances in nephrology and dramatic decreases in the incidence of cardiovascular disease [1], the number of patients beginning renal replacement therapy is annually increasing in Japan as well as in Western countries [2, 3]. Major causes of end-stage renal disease (ESRD) are diabetes and hypertension; however, the effects of other cardiovascular risk factors on the human kidney are not well understood. Further clarification of the pathogenesis and risk factors of renal histologic changes in the general population might provide useful information for preventing chronic renal diseases that tend to be asymptomatic and often go undiagnosed. However, most of the reported findings concerning this issue have come from animal models [4–7] or selected patients [8–10]; little information has been made available with respect to the general population, due to various methodologic obstacles in etiologic research on renal diseases [11].

A prospective population-based study of cardiovascular disease has been carried out since 1961 in Hisayama Town on Kyushu Island in southern Japan. The most characteristic feature of this study is that the cause of death has been verified by autopsy in 80% of the deceased subjects from the study population [12–15]. A previous report of 270 autopsies of Hisayama residents [12] showed that both age and hypertension were closely related to the reduction in kidney weight, as well as the progression of glomerular sclerosis and nephrosclerosis. However, it did not assess the effects of other cardiovascular risk factors. The Honolulu Heart Program [16, 17] is, to our knowledge, the only other population-based study that has examined this issue, although the autopsy rate in this study was low (20.6%). In the present study, we examined renal histologic changes in most of the deceased Hisayama residents and showed that various cardiovascular risk factors were associated with the development of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis.

METHODS

Study population

The population of Hisayama Town is approximately 7500 and has been shown to be representative of Japan as a whole based on data from the national census [12, 13]. The study design and characteristics of the subject population have been described in detail elsewhere [14, 15]. From January 1962 to December 1994, a total of 1742 Hisayama residents of all age groups died, and of these, 1394 (80.0%) underwent autopsy examinations. Autopsy rate was not different between men (78.7%) and women (81.6%). Among these consecutive autopsy subjects, 1168 participated in at least one of the six health examinations in 1961, 1967, 1974, 1978, 1983, and 1988. In every examination, the participation rate exceeded more than 80% of all the Hisayama residents 40 years old or older. We excluded 98 subjects who were missing the preserved renal tissues, 33 subjects with degenerated or small renal tissues, 80 subjects who underwent autopsy examination in other hospitals, and 118 subjects who had had no recent health examination data before death. Finally, 839 subjects with adequate renal tissues and health examination data just before death (mean period, 3.5 ± 1.8 years; range, 0 to 7 years) were enrolled in the present study.

Morphologic examination of renal tissue

For light microscopic study, paraffin-embedded renal tissues obtained by standard autopsy methods were cut at 2 μm thickness and stained with periodic acid-Schiff (PAS). The semiquantitative score according to the method of Raij, Azar and Keane [18] was used to evaluate the degree of glomerular sclerosis. For each tissue specimen, 100 glomeruli from the superficial to deep cortex were examined uniformly, and the severity of the lesion in each glomerulus was graded from 0 to 4+ according to the percentage of glomerular sclerosis. Specifically, a score of 0 represented a complete absence of sclerotic lesion of the glomerulus, 1+ represented 1% to 25% involvement of sclerotic lesion of the glomerulus, and 2+, 3+, and 4+ represented 26% to 50%, 51% to 75%, and 76% to 100% involvement of the glomerulus, respectively. An injury score was then obtained by multiplying the degree of damage (0 to 4+) by the number of glomeruli with the same degree of injury. That is, the glomerular sclerosis index was calculated by the following formula:

$$\text{Glomerular sclerosis index} = \frac{n_0 \times 0 + n_1 \times 1 + n_2 \times 2 + n_3 \times 3 + n_4 \times 4}{4}$$

The variables n_0, n_1, n_2, n_3, and n_4 indicate the number of glomeruli showing sclerotic lesion scores of 0 to 4+, respectively.

The degree of arteriolar hyalinosis was assessed semiquantitatively by the method of Bader and Meyer [19]. For each tissue specimen, 50 arterioles were examined and the severity of the lesion in each arteriole was graded from 1+ to 4+ according to the extent of arteriolar hyalinosis as follows: 1+ represented the absence of any conspicuous alteration of the arteriolar wall, 2+ represented arteriolar wall hyalinosis comprising less than 50% of the arteriolar circumference, 3+ represented arteriolar wall hyalinosis of more than 50% but less than 100% of the arteriolar circumference, and 4+ represented hyalinization of the entire arteriolar wall. The arteriolar hyalinosis index was calculated by the following formula:

$$\text{Arteriolar hyalinosis index} = \frac{n_1 \times 1 + n_2 \times 2 + n_3 \times 3 + n_4 \times 4}{50}$$

Here, n_1, n_2, n_3, and n_4 indicate the number of arterioles showing hyalinosis scores of 1+ to 4+, respectively.

The wall-lumen ratio was evaluated as the severity of arteriosclerosis by the method of Kernohan, Anderson, and Keith [20]. For each tissue specimen, all arteries with an outer diameter exceeding 60 μm were examined using an eyepiece micrometer. The outer diameter and the lumen diameter of least axis of the elliptic profile were directly measured. The wall-lumen ratio was calculated in each artery as lumen diameter/(outer diameter − lumen diameter)/2, and the mean value among all arteries in each subject was used as the index of arteriosclerosis. Because the wall-lumen ratio differs by arterial size, we further examined the degree of arteriosclerosis by classifying arteries into four categories according to their size. However, because the frequencies and risk profiles for all these categories were highly similar, we showed only the results of all arteries together in the present study. Renal tubulointerstitial changes were not examined in this study, since many subjects underwent autopsy examination more than 24 hours after death. All histologic evaluations were carried out by one of the authors (M.K.) with no information other than the serial autopsy number.

To differentiate the effect of cardiovascular risk factors from age-related changes, we selected 103 subjects who had none of the following characteristics: proteinuria, hematuria, renal failure (creatinine clearance ≤0.5 mL/second as estimated by the Cockcroft-Gault formula), hypertension, glucose intolerance, or primary renal disease at autopsy. Using this subgroup, the cut-off limits were drawn from the upper 95th percentile or the lower 5th percentile of these histologic parameter distributions; that is, the development of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis were defined as a glomerular sclerosis index >20, an arteriolar hyalinosis index >1.56, and a wall-lumen ratio <1.30, respectively.

Risk factors

Blood pressures were measured three times using a standard mercury sphygmomanometer at every examination, and the mean values were used for the analysis. Hypertension was defined as systolic blood pressure ≥140 mm Hg and/or diastolic blood pressure ≥90 mm Hg and/or the current use of antihypertensive agents. Glucose intolerance was defined by an oral glucose tolerance test in the subjects with glycosuria in 1961 and 1967, by fasting and postprandial glucose concentrations in 1974, 1978, and 1983, and by a 75 g oral glucose tolerance test in 1988, in addition to medical history of diabetes. Electrocardiogram (ECG) was recorded at every examination, and ECG abnormalities were defined as Minnesota code 3-1 and/or 4-1,2,3. Serum cholesterol levels were measured by the Zak-Henly method with a modification by Yoshikawa in 1961 and 1967, by the Zurkowski method in 1974, and by the enzymatic method after 1978. Serum creatinine concentration was measured by Jaffe's method after 1974, and glomerular filtration rate was calculated by the Modification of Diet in Renal Disease (MDRD) Study Group formula [21]. Freshly voided urine samples were tested by the sulfosalicylic acid method in 1961 and 1967, by the dipstick method after 1974, and proteinuria and hematuria were defined as 1+ or more. Body height and weight were measured in light clothing without shoes and the body mass index (kg/m^2) was calculated. Information on antihypertensive treatment, alcohol intake, and smoking habits was obtained by means of a standard questionnaire, and classified as current habitual use or a lack thereof.

Statistical analysis

Mean values and frequencies of variables were compared using Student t test and chi-square test as appropriate. A logistic regression model was applied to identify the effect of cardiovascular risk factors on the development of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis, and the odds ratio (OR) and 95% confidence interval (CI) were calculated. In multivariate analysis, only significant variables obtained in the age-adjusted analysis were used. Men and women were separately examined in all analyses. Levels of $P < 0.05$ were considered to indicate statistical significance.

RESULTS

Table 1 summarizes the characteristics of the 839 autopsy subjects at the health examinations by gender. Women were approximately 5 years older on average at death than men. Men had higher serum creatinine level than women, while mean glomerular filtration rate was higher in women. Women had higher mean systolic blood pressure and pulse pressure than men, but there was no gender difference in mean diastolic blood pressure and mean arterial pressure. The frequencies of glucose intolerance, alcohol intake, and smoking habits were higher in men, while the mean total cholesterol level was higher in women. Mean body mass index level and the frequencies of proteinuria, hematuria, antihypertensive treatment, and ECG abnormalities were not different between men and women.

Table 1. Characteristics of the 839 autopsy subjects at health examinations before death by sex, The Hisayama Study

Variables	Men (N = 458)	Women (N = 381)
Age at death *years*	73 ± 12	78 ± 11[a]
Serum creatinine *mmol/L*	97 ± 29	87 ± 50[a]
Glomerular filtration rate *mL/min/1.73 m^2*	75.6 ± 19.7	87.1 ± 23.8[a]
Proteinuria %	13.9	16.7
Hematuria %	5.3	6.0
Systolic blood pressure *mm Hg*	147 ± 29	154 ± 28[a]
Diastolic blood pressure *mm Hg*	81 ± 14	80 ± 14
Mean arterial pressure *mm Hg*	103 ± 17	104 ± 17
Pulse pressure *mm Hg*	66 ± 23	74 ± 23[a]
Antihypertensive agents %	20.5	23.0
Glucose intolerance %	27.1	16.5[a]
ECG abnormalities %	25.0	29.8
Total cholesterol *mmol/L*	4.5 ± 1.1	4.9 ± 1.2[a]
Body mass index *kg/m^2*	20.7 ± 2.8	21.0 ± 3.6
Alcohol intake %	50.1	6.6[a]
Smoking habits %	59.0	14.2[a]

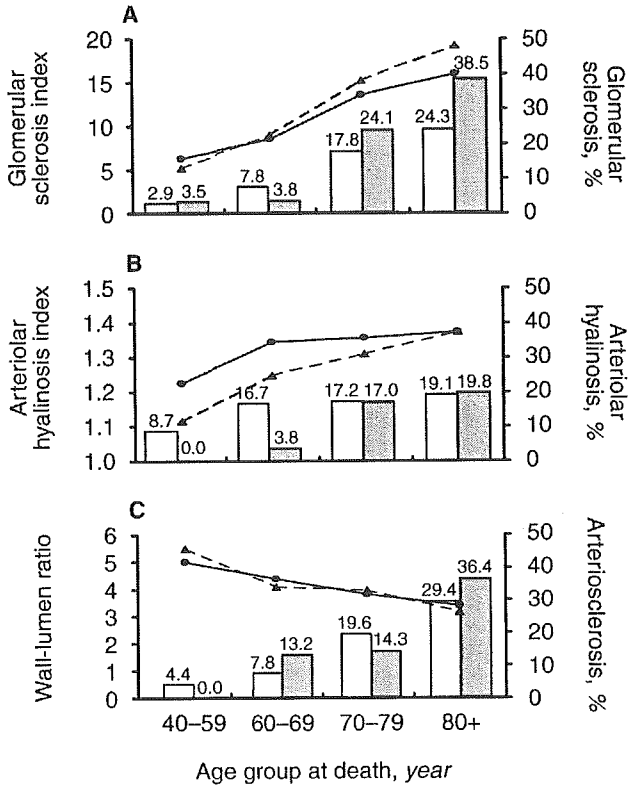
Glomerular filtration rate determined by Modification of Diet in Renal Disease (MDRD) Study Group formula [21]. Serum creatinine, glomerular filtration rate, and hematuria were measured in 329 men and 270 women who died after 1974. Values are expressed as mean ± SD or percentage.

[a] $P < 0.01$ vs. men

The age-specific mean values of renal histologic parameters are shown by gender in Figure 1. With elevating age, the mean values of the glomerular sclerosis index and arteriolar hyalinosis index linearly increased, and that of the wall-lumen ratio linearly decreased in both men and women.

The frequencies of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis were 15.5%, 16.4%, and 18.1% in men, respectively, and 26.8%, 15.2% and 23.9% in women, respectively. The frequency of glomerular sclerosis was 2.9% for men and 3.5% for women in the 40- to 59-year-old age group, and significantly increased to 24.3% and 38.5% in subjects 80 years old or older, respectively (Fig. 1). Likewise, the frequency of arteriolar hyalinosis significantly increased from 8.7% to 19.1% for men and from 0% to 19.8% for women. A similar pattern was observed for arteriosclerosis; the frequency increased from 4.4% to 29.4% for men and from 0% to 36.4% for women. These associations of glomerular and renal vascular changes with age were similar for both men and women.

We estimated the age-adjusted ORs and 95% CIs of each cardiovascular risk factor for the development of glomerular and vascular changes by gender (Tables 2 and 3). Serum creatinine, glomerular filtration rate, and



Number of subjects				
Men	69	90	163	136
Women	29	53	112	187

Fig. 1. Age-specific mean values and frequencies of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis among the 839 autopsy subjects by gender [men (□) (●) and women (▨) (▲)], The Hisayama Study, 1962-1994. Solid and dashed lines indicate age-specific mean values of glomerular sclerosis index, arteriolar hyalinosis index, and wall-lumen ratio in men and women, respectively.

proteinuria significantly associated with the risk of glomerular sclerosis in both men and women. Systolic blood pressure and pulse pressure significantly increased the risk of glomerular sclerosis in both men and women, and mean arterial pressure increased the risk only in women. Pulse pressure remained a significant risk factor for glomerular sclerosis after being adjusted for age and mean arterial pressure in both men and women (OR, 1.17; 95% CI, 1.03 to 1.33 for men; and OR, 1.25; 95% CI, 1.09 to 1.43 for women). In addition, glucose intolerance was found to be a significant risk factor in men, as were ECG abnormalities in women. Alcohol intake had a significant protective effect on glomerular sclerosis in women.

For arteriolar hyalinosis, higher serum creatinine level for men and lower glomerular filtration rate for women were significant risk factors. Proteinuria significantly increased the risk of arterial hyalinosis in both men and women. Systolic, diastolic, mean, and pulse pressures were all significant risk factors in both men and women. After adjustment of age and mean arterial pressure, pulse pressure was a significant risk factor for arterial hyalinosis in men (OR, 1.17; 95% CI, 1.02 to 1.34), but was not in women (OR, 1.06; 95% CI, 0.91 to 1.23). Glucose intolerance, ECG abnormalities and total cholesterol levels were additional risk factors in men. For arteriosclerosis, glomerular filtration rate and proteinuria were significant risk factors in both men and women. Systolic blood pressure, mean arterial pressure, and pulse pressure were found to be significant risk factors in both men and women, as was diastolic blood pressure in women. After adjusted for age and mean arterial pressure, pulse pressure remained a significant risk factor for both men and women (OR, 1.28; 95% CI, 1.11 to 1.47 for men; and OR, 1.14; 95% CI, 1.00 to 1.31 for women). In addition, ECG abnormalities significantly increased the risk of arteriosclerosis in both men and women, and alcohol intake had a significant protective effect in women.

Results of the multivariate analysis of risk factors for the development of glomerular and renal vascular changes are summarized by gender in Table 4. We did not include serum creatinine and glomerular filtration rate in the multivariate analysis, since these parameters were not the cause but the result of renal glomerular and vascular changes. Both age and systolic blood pressure were significant independent risk factors for each of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis in both men and women. In addition, glucose intolerance and proteinuria for men were found to be significant risk factors for glomerular sclerosis. Elevated total cholesterol level significantly increased the risk of arteriolar hyalinosis in men. ECG abnormalities were an independent risk factor for arteriosclerosis in both men and women, and proteinuria was an additional risk factor in women. Alcohol intake tended to have a protective effect on glomerular sclerosis ($P = 0.07$) and arteriosclerosis ($P = 0.06$) in women. When using mean arterial pressure instead of systolic blood pressure in the multivariate model, mean arterial pressure similarly increased the risk for glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis for both men and women (data not shown). In the multivariate analysis, including mean arterial pressure and pulse pressure simultaneously, the latter was a significant risk factor for glomerular sclerosis (OR, 1.23; 95% CI, 1.00 to 1.33), arteriolar hyalinosis (OR, 1.19; 95% CI, 1.01 to 1.39), and arteriosclerosis (OR, 1.23; 95% CI, 1.05 to 1.45) for men. Likewise, for women, pulse pressure was also a significant risk factor for glomerular sclerosis (OR, 1.32; 95% CI, 1.13 to 1.54), but not for arteriolar hyalinosis (OR, 1.06; 95% CI, 0.90 to1.24), or arteriosclerosis (OR, 1.11; 95% CI, 0.96 to 1.29). Additional multivariate models were fit to explore the possi-

Table 2. Age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) of risk factors for the development of glomerular and vascular changes among 458 men autopsy subjects, The Hisayama Study, 1962 to 1994

	Glomerular sclerosis		Arteriolar hyalinosis		Arteriosclerosis	
Risk factor	OR	95% CI	OR	95% CI	OR	95% CI
Serum creatinine (10 mmol/L)	1.41[a]	1.22–1.63	1.12[b]	1.01–1.25	1.48[a]	1.27–1.72
Glomerular filtration rate (10 mL/min/1.73 m^2)	0.58[a]	0.47–0.73	0.87	0.73–1.03	0.54[a]	0.43–0.68
Proteinuria (Yes/no)	4.10[a]	2.21–7.63	2.52[a]	1.35–4.72	2.41[a]	1.28–4.54
Hematuria (Yes/no)	0.53	0.11–2.44	0.54	0.12–2.46	0.66	0.17–2.49
Systolic blood pressure (10 mm Hg)	1.11[b]	1.02–1.21	1.25[a]	1.15–1.37	1.38[a]	1.25–1.52
Diastolic blood pressure (10 mm Hg)	1.03	0.86–1.24	1.36[a]	1.12–1.64	1.49[a]	1.23–1.82
Mean arterial pressure (10 mm Hg)	1.12	0.93–1.30	1.41[a]	1.21–1.64	1.62[a]	1.38–1.91
Pulse pressure (10 mm Hg)	1.17[a]	1.05–1.30	1.28[a]	1.15–1.43	1.42[a]	1.27–1.60
Glucose intolerance (Yes/no)	2.43[a]	1.41–4.18	2.06[a]	1.23–3.46	1.30	0.75–2.25
Electrocardiogram abnormalities (Yes/no)	1.19	0.66–2.16	1.82[b]	1.07–3.12	4.62[a]	2.66–8.01
Total cholesterol (Yes/no)	1.01	0.79–1.29	1.50[a]	1.19–1.88	1.16	0.92–1.47
Body mass index (1 kg/m^2)	0.99	0.90–1.09	1.01	0.92–1.11	1.04	0.95–1.15
Alcohol intake (Yes/no)	0.72	0.42–1.24	0.66	0.39–1.11	0.85	0.51–1.41
Smoking habits (Yes/no)	0.75	0.44–1.28	1.09	0.65–1.82	1.08	0.64–1.81

Odds ratios were calculated for the increment in parentheses. Risk of serum creatinine, glomerular filtration rate, and hematuria were estimated in 329 men who died after 1974.
[a]$P < 0.01$; [b]$P < 0.05$

Table 3. Age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) of risk factors for the development of glomerular and vascular changes among 381 women autopsy subjects, The Hisayama Study, 1962 to 1994

	Glomerular sclerosis		Arteriolar hyalinosis		Arteriosclerosis	
Risk factor	OR	95% CI	OR	95% CI	OR	95% CI
Serum creatinine (10 mmol/L)	1.12[a]	1.01–1.23	1.03	0.97–1.08	1.07	0.98–1.16
Glomerular filtration rate (10 mL/min/1.73 m^2)	0.71[b]	0.61–0.82	0.85[a]	0.73–0.99	0.73[b]	0.63–0.84
Proteinuria (Yes/no)	2.53[b]	1.35–4.74	2.41[a]	1.20–4.83	3.55[b]	1.89–6.64
Hematuria (Yes/no)	0.43	0.11–1.65	NA		0.54	0.14–2.04
Systolic blood pressure (10 mm Hg)	1.20[b]	1.10–1.32	1.18[b]	1.06–1.30	1.16[b]	1.06–1.26
Diastolic blood pressure (10 mm Hg)	1.11	0.93–1.33	1.30[a]	1.06–1.60	1.14	0.95–1.37
Mean arterial pressure (10 mm Hg)	1.25[b]	1.08–1.45	1.32[b]	1.11–1.57	1.22[b]	1.05–1.42
Pulse pressure (10 mm Hg)	1.28[b]	1.14–1.43	1.17[a]	1.03–1.32	1.19[b]	1.07–1.33
Glucose intolerance (Yes/no)	1.60	0.87–2.94	1.63	0.81–3.26	1.05	0.54–2.02
Electrocardiogram abnormalities (Yes/no)	1.85[a]	1.11–3.09	1.52	0.84–2.75	3.03[b]	1.79–5.14
Total cholesterol (Yes/no)	1.16	0.95–1.42	1.08	0.85–1.36	1.08	0.87–1.33
Body mass index (1 kg/m^2)	1.01	0.94–1.08	1.04	0.96–1.13	0.93	0.86–1.00
Alcohol intake (Yes/no)	0.21[a]	0.05–0.92	1.94	0.73–5.18	0.11[a]	0.01–0.87
Smoking habits (Yes/no)	0.61	0.29–1.31	1.64	0.78–3.44	0.45	0.19–1.05

NA, Not available. Odds ratios were calculated for the increment in parentheses. Risk of serum creatinine, glomerular filtration rate, and hematuria were estimated in 270 women who died after 1974.
[a]$P < 0.05$; [b]$P < 0.01$

bility of interactions between the variables in the model, but no such interaction was identified. There was no evidence of a lack of fit in the multivariate model composed of significant risk factors.

To examine the combined effect of blood pressure and glucose intolerance, we stratified the subjects into four groups according to hypertension and glucose intolerance status (Table 5). The age-adjusted analysis showed that hypertension alone significantly increased the risk of glomerular sclerosis, arteriolar hyalinosis, and arteriosclerosis in both men and women, while glucose intolerance alone did not. When glucose intolerance was combined with hypertension, the age-adjusted ORs for glomerular and vascular changes further increased in both men and women, but this additive effect was modest for arteriosclerosis.

In 187 subjects with glucose intolerance, the age- and gender-adjusted analysis showed similar results as those of the whole subjects. In the multivariate analysis, age and proteinuria were significant independent risk factors for glomerular sclerosis, and alcohol intake had a protective effect. Likewise, systolic blood pressure and total cholesterol level were found to be independent risk factors for arteriolar hyalinosis. Age, ECG abnormalities, and proteinuria significantly increased the risk of arteriosclerosis.

DISCUSSION

In this autopsy-based population survey, we histopathologically examined glomerular sclerosis, renal arteriolar hyalinosis, and arteriosclerosis, and analyzed the effects

Table 4. Multivariate odds ratios (ORs) and 95% confidence intervals (CIs) of risk factors for the development of glomerular and renal vascular changes among the 839 autopsy subjects by gender, The Hisayama Study, 1962 to 1994

Risk factor	Glomerular sclerosis		Arteriolar hyalinosis		Arteriosclerosis	
	OR	95% CI	OR	95% CI	OR	95% CI
Men (N = 458)						
Age at death (10 years)	1.76[a]	1.32–2.34	1.14	0.88–1.46	2.19[a]	1.59–3.02
Systolic blood pressure (10 mm Hg)	1.07	0.97–1.17	1.23[a]	1.11–1.37	1.31[a]	1.18–1.46
Glucose intolerance (Yes/no)	2.29[a]	1.28–4.09	1.72	0.95–3.10		
Electrocardiogram abnormalities (Yes/no)			1.25	0.66–2.37	2.78[a]	1.51–5.13
Total cholesterol (1 mmol/L)			1.57[a]	1.21–2.03		
Proteinuria (Yes/no)	3.45[a]	1.80–6.59	1.65	0.81–3.34	1.69	0.84–3.43
Women (N = 381)						
Age at death (10 years)	2.11[a]	1.55–2.88	1.45[b]	1.05–2.00	2.12[a]	1.54–2.91
Systolic blood pressure (10 mm Hg)	1.16[a]	1.04–1.30	1.15[b]	1.02–1.30	1.04	0.93–1.17
Electrocardiogram abnormalities (Yes/no)	1.36	0.78–2.39			2.41[a]	1.36–4.25
Alcohol intake (Yes/no)	0.24	0.05–1.15			0.14	0.02–1.13
Proteinuria (Yes/no)	1.63	0.80–3.30	1.63	0.75–3.55	2.81[a]	1.39–5.69

Age and all significant risk factors available in the age-adjusted analysis were included in the multivariate model. Odds ratios were calculated for the increment in parentheses.
[a]$P < 0.01$; [b]$P < 0.05$

Table 5. Age-adjusted odds ratios (ORs) and 95% confidence intervals (CIs) for the development of glomerular and renal vascular changes according to hypertension (HT) and glucose intolerance (GI) status by gender, The Hisayama Study, 1962 to 1994

Category	Number of patients	Glomerular sclerosis		Arteriolar hyalinosis		Arteriosclerosis	
		OR	95% CI	OR	95% CI	OR	95% CI
Men (N = 458)							
No HT and No GI	132	1.00		1.00		1.00	
GI alone	38	2.50	0.81–7.69	2.85	0.92–8.80	1.49	0.36–6.25
HT alone	202	2.12	0.96–4.66	3.29[a]	1.47–7.36	4.94[a]	2.12–11.51
HT with GI	86	4.89[a]	2.11–11.32	5.78[a]	2.45–13.65	5.83[a]	2.30–14.75
Women (N = 381)							
No HT and No GI	98	1.00		1.00		1.00	
GI alone	15	0.99	0.19–5.17	1.65	0.17–15.96	1.21	0.23–6.37
HT alone	220	2.31[b]	1.17–4.54	4.66[a]	1.61–13.51	2.66[a]	1.30–5.42
HT with GI	48	3.83[a]	1.63–9.01	7.08[a]	2.13–23.54	2.55[b]	1.02–6.40

[a]$P < 0.01$; [b]$P < 0.05$

of cardiovascular risk factors on these renal pathologic changes using a logistic regression model. We confirmed that age and systolic blood pressure were common risk factors for all these histopathologic changes. In addition, metabolic abnormalities, that is, glucose intolerance and elevated serum cholesterol, and ECG abnormalities affected glomerular and vascular changes, while alcohol intake had a protective effect.

Age

Our study showed that the frequencies of each of glomerular sclerosis, hyaline arteriosclerosis, and arteriosclerosis linearly increased with advancing age in both men and women, and these associations were independent of other risk factors. It is well known that both glomerular filtration rate and renal blood flow progressively decline with age [5]. These findings, together with those of the present study, suggest that aging itself might induce the progression of glomerular sclerosis and arteriosclerosis, ultimately resulting in an age-related deterioration of renal function in the general population.

Hypertension

Hypertension is one of the major risk factors for development of ESRD [22, 23]. We showed that elevated blood pressure was a significant independent risk factor, not only for the development of arteriolar hyalinosis and arteriosclerosis, but also for glomerular sclerosis, even after adjustment for age and other risk factors. The impact of mean arterial pressure and pulse pressure on glomerular and vascular changes was similar to that of systolic blood pressure, and the impact of pulse pressure remained significant even after controlling for mean arterial pressure. Moreover, the effect of blood pressure was stronger for arteriolar hyalinosis and arteriosclerosis than glomerular sclerosis. Essential hypertension, the main type of hypertension seen in this study, is considered to cause arteriolar hyalinosis and arteriosclerosis, resulting in the progression of glomerular sclerosis [24].

Glucose intolerance

Diabetes is well known to cause diabetic nephropathy and accelerated atherosclerosis. In this study, glucose

intolerance was a significant risk factor for the development of glomerular sclerosis in men. When the combined effect of hypertension and glucose intolerance was examined, the risk of glomerular and vascular changes was mainly determined by hypertension, and glucose intolerance only had an additive effect, especially in women. Diabetic nephropathy is known to be more prevalent and to progress more rapidly in men than in women [6]. Moreover, in our cohort, the frequency of glucose intolerance was higher in men. These factors may have been responsible for the present finding that glucose intolerance was much more highly associated in men. In contrast, arteriosclerotic vascular change is generally a slow pathoanatomic process that may require a long period of time for progression. Our study subjects underwent autopsy within 7 years after their last health examination, and thus the findings of the present study might reflect only short-term effects. This, in turn, may have resulted in the finding that glucose intolerance was not a risk factor for renal vascular changes.

ECG abnormalities

Our data showed that ECG abnormalities were a significant independent risk factor for renal arteriosclerosis in both men and women. In our subjects, the frequency of ECG abnormalities increased with elevating age and blood pressure (data not shown), suggesting that these factors reflect a longer duration of hypertension. Another explanation is that renal arteriosclerosis might be closely related to systemic atherosclerosis, including that of coronary atherosclerosis. An autopsy study by the Honolulu Heart Program also found that the frequency of cardiovascular death and the degree of aortic atherosclerosis linearly increased with the progression of renal arteriosclerosis [16].

Total cholesterol

Experimental studies suggest that circulating lipoproteins play a significant role in the pathogenesis of glomerular sclerosis [25]. However, there have been no available data concerning the effect of hyperlipidemia on renal histologic changes in the general population. Our data showed that total cholesterol level was significantly related to the development of arteriolar hyalinosis in men. This finding is consistent with those of Tracy et al [8], who studied autopsies from a hospital and coroner's office. On the other hand, the total cholesterol level in our subjects was lower than that of Western populations, and this might be a reason for the lack of association between serum cholesterol level and arteriosclerosis in this study.

Alcohol intake

There has been little information concerning the effect of alcohol intake on renal histologic changes. The Honolulu Heart Program reported a protective association between alcohol intake and arteriolar hyalinosis [17], but did not examine the effect of alcohol intake on glomerular sclerosis and arteriosclerosis. Our data first showed that alcohol intake has a protective effect on the development of glomerular sclerosis and arteriosclerosis in women. The majority of our women drinkers consumed a small amount of alcohol [14], suggesting that light alcohol consumption might protect against the progression of glomerular sclerosis and arteriosclerosis. This finding is consistent with the fact that light-to-moderate alcohol consumption significantly reduces the risk of cardiovascular disease [14]. The protective effect of alcohol is thought to be mediated by the beneficial effect of alcohol on high-density lipoprotein cholesterol level [26].

Validation study

A previous report of Tracy et al [27] claimed that mean arterial pressure and age could be used to calculate interlobular artery wall thickness with great precision. Wall thickness (%) could be calculated from $0.171 \times$ mean arterial pressure $+ 0.047 \times$ age $+ 1.1$ and $0.140 \times$ mean arterial pressure $+ 0.092 \times$ age $+ 4.0$, for arteries of sizes of 80 to 150 μm and 150 to 300 μm, respectively. To evaluate the efficacy of this formula by Tracy et al, we performed a validation study using our study subjects. Mean wall thickness calculated as wall thickness/outer diameter, being $22.1\% \pm 5.2\%$ in our study, was not different from that of $22.3\% \pm 3.0\%$ by the Tracy et al formula for arteries of sizes of 150 to 300 μm, but it was significantly lower in our study ($18.3\% \pm 4.5\%$) than by the formula of Tracy et al ($25.4\% \pm 2.7\%$) for arteries of sizes of 80 to 150 μm. Since many cardiovascular risk factors affect renal arteriosclerosis, the formula of Tracy et al might have a poor predictable value for estimating renal arteriosclerosis of small artery.

Limitations of the study

We used the wall-lumen ratio as an index of arteriosclerosis. Arteries in autopsy tissues are collapsed, leading to a low wall-lumen ratio. This might result in overestimation of the degree of arteriosclerosis, when tissues are obtained without perfusion-fixation. Tracy, Heigle, and Velez-Duran [9] examined this problem using autopsy subjects in whom one kidney was perfusion-fixed and the other immersion-fixed. They showed that the outer diameter of immersion-fixed vessels was reduced and the wall thickness extended in the same proportion as in perfusion-fixed vessels, regardless of the vessel size. The wall-lumen ratio in each artery might be overestimated to the same degree. In the present study, since the development of arteriosclerosis was determined by the lower 5th percentile of the wall-lumen ratio, postmortem collapse of arteries was not likely to have distorted the findings.

Renal tubulointerstitial change is one of the causes of renal functional decline in animal models or patients with various renal diseases [4, 7]. However, we did not examine this pathologic change, since many subjects in our study underwent autopsy examination more than 24 hours after death, and their postmortem interstitial changes were severe. A previous autopsy study found no significant association between interstitial fibrosis and risk factors such as age and blood pressure, probably due to postmortem edema [10]. Thus, information on interstitial changes might not have substantially improved the value of our findings.

A consideration of great importance in any epidemiologic use of autopsies is the special subset of cases that entered the autopsy series by dying of conditions unrelated to cardiovascular diseases [28, 29]. These include cancers, infections, violence, and many other conditions. However, all of the glomerular and vascular changes in these subjects also similarly increased with age as those of the whole study subjects (data not shown). The Hisayama Study encourages performance of autopsy examinations in all of the deceased Hisayama residents, regardless of the cause of death, and maintains high autopsy rate throughout the study period. Therefore, the results of our findings might have generalizability. Because this study is based on autopsies, the subjects in this study may have shown a higher prevalence of risk factors and more severe renal histologic changes than the overall population. This selection bias might affect their associations with risk factors. Despite these limitations, we believe that the findings of this study provide useful information toward a better understanding of the pathogenesis of kidney damage in the general population.

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Validity of the JNC VI Recommendations for the Management of Hypertension in a General Population of Japanese Elderly

The Hisayama Study

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Background: It is not known whether the treatment recommendations presented in the Sixth Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure are applicable to the Japanese elderly population.

Methods: We followed up 588 cardiovascular disease–free residents of a Japanese community who were 60 years or older from November 1, 1961, through October 31, 1993. Treated hypertensive patients were excluded from the analysis. During this period, CVD occurred in 179 subjects. The incidences were estimated by the pooling of repeated observations method.

Results: The age- and sex-adjusted incidences of cardiovascular disease significantly increased with elevated blood pressure levels. The hazard ratio for stage 3 hypertension was 5.34 (95% confidence interval, 2.66-10.71; $P<.001$) compared with optimal blood pressure after adjustment for other covariates. Among subjects aged 60 to 79 years, the incidences for stages 1 through 3 hypertension were significantly higher than for those with optimal and normal blood pressure. In comparison, among those 80 years or older, the incidence was significantly higher only in patients with stage 3 hypertension. We further estimated the incidences according to the risk stratification system. In the younger elderly subjects, the incidences increased with rising blood pressure levels in each risk stratum. Similar relationships were not observed among the older elderly subjects.

Conclusions: Our findings demonstrate that the recommendations of the Sixth Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure were potentially applicable to the Japanese elderly subjects 79 years or younger. Based on our findings, however, hypertension might not be a risk factor for cardiovascular disease among very old hypertensive patients with advanced atherosclerosis.

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THE SIXTH Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC VI) recommended consideration of patient-specific estimates of absolute (rather than relative) risks for cardiovascular disease (CVD) in treatment decisions.[1] For the first time, the JNC VI proposed a risk stratification system that was based not only on the level of blood pressure (BP) but also on the presence or absence of target organ damage (TOD) or other risk factors such as smoking, dyslipidemia, and diabetes. A prospective cohort study of National Health and Nutrition Examination Survey I demonstrated an absolute benefit derived from treating hypertension according to this risk stratification system in the US population.[2] It is unknown whether this system works well in the Japanese elderly population.

The therapeutic guidelines of the Research Group for Long-term Prognosis of Hypertension in the Elderly were published in Japan in 1999.[3] The JNC VI[1] and Japanese guidelines[3] are generally in accord with regard to the principles of drug prescription. However, a serious discrepancy exists between these 2 sets of guidelines on the level of BP at which antihypertensive treatment should be initiated. Among patients 70 years or older, the systolic BP levels at which antihypertensive treatment is recommended are 20 to 40 mm Hg higher in the Japanese guidelines[3] than in the JNC VI.[1] It is not clear which recommendation is better for the Japanese elderly population.

To clarify this issue, we examined the contribution of hypertension to risk for

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CVD occurrence in a general population of Japanese elderly subjects not taking antihypertensive medication and evaluated whether the JNC VI treatment recommendations were applicable.

METHODS

STUDY DESIGN

The Hisayama Study is an ongoing population-based epidemiological study designed to investigate the morbidity and mortality of CVD and its risk factors in the town of Hisayama, Japan.[4-9] At the initial screening in 1961, 588 subjects 60 years or older, all of whom were free of CVD, were registered as a cohort population, which included almost 90% of the total population of this age group. Subsequent examinations were conducted in 1967, 1974, 1978, 1983, and 1988. The response rates for these examinations were 91.8%, 90.9%, 85.9%, 74.2%, and 81.4% of CVD-free survivors, respectively. Because this report focuses on the incidence of CVD in subjects who were not taking antihypertensive medication, we excluded treated hypertensive patients from the analysis at each examination (22, 82, 45, 27, 12, and 9 subjects, respectively).

At each examination, we collected medical and life histories, conducted a physical examination and urinalysis for protein and sugar levels, measured levels of serum cholesterol, and performed electrocardiography (ECG). Information on antihypertensive treatment, smoking habits, and alcohol intake was obtained using a standard questionnaire, and these factors were classified as habitual or not. Blood pressure was measured 3 times with the subject in the recumbent position, after having rested for at least 5 minutes, by means of the standard sphygmomanometer with a standard cuff. Korotkoff phase 5 was taken as the diastolic BP unless the sounds persisted at 0, in which case Korotkoff phase 4 was recorded. The mean of 3 measurements was used in the present analysis. Body weight and height were measured with the subject in light clothing without shoes, and body mass index (calculated as weight in kilograms divided by the square of height in meters) was determined. Serum cholesterol level was determined by the Zak-Henly method, including a modification by Yoshikawa, in 1961 and 1967; by the Zurkowski method in 1974; and by the enzymatic method in 1978, 1983, and 1988.[5,10] Glucose intolerance was determined by means of an oral glucose tolerance test in subjects with glycosuria in 1961 and 1967; by means of fasting and postprandial glucose concentrations in 1974, 1978, and 1983; and by means of a 75-g oral glucose tolerance test in 1988, in addition to the medical history of diabetes.[7,8] Electrocardiographic abnormalities were determined as Minnesota codes 3-1 (high R-waves), 4-1, 4-2, and/or 4-3 (ST-segment depression).

BP CLASSIFICATION AND RISK STRATIFICATION

The JNC VI recommended treating hypertension according to a risk stratification system based on BP categories and risk groups (A-C).[1] We used the following BP categories: optimal (systolic BP [SBP], <120 mm Hg and diastolic BP [DBP], <80 mm Hg), normal (SBP, 120-129 or DBP, 80-84), high-normal (SBP, 130-139 or DBP, 85-89), stage 1 (SBP, 140-159 or DBP, 90-99), stage 2 (SBP, 160-179 or DBP, 100-109), and stage 3 (SBP, ≥180 or DBP, ≥110). In our study, risk group B included patients who did not have TOD or glucose intolerance, but had 1 or more risk factors, such as 60 years or older, sex (men and postmenopausal women), dyslipidemia, or a smoking habit. Risk group C included subjects who had TOD such as ECG abnormalities and proteinuria, or glucose intolerance. Since our subjects had at least 2 risk factors (age and sex), all of them were categorized in risk group B or C.

END-POINT DEFINITION AND ASCERTAINMENT

The subjects were followed up prospectively from November 1, 1961, through October 31, 1993, by means of repeated health examinations or a daily monitoring system established by the study team and local physicians or by members of the local health and welfare office. During this period, no subjects were lost to follow-up.

Cardiovascular disease included all types of stroke and coronary heart disease (CHD). The clinical diagnosis of stroke was determined on the basis of a detailed history, results of a neurological examination, and computed tomographic findings.[9] The diagnosis of CHD included acute or silent myocardial infarction and sudden cardiac death within 1 hour after the onset of acute illness. The diagnosis of CHD was based on clinical symptoms and results of ancillary diagnostic procedures, including ECG recordings, cardiac enzyme levels, echocardiography, or coronary angiography. During the follow-up period, 471 subjects who had not been taking antihypertensive medication died, and autopsies were performed on 373 (79.2%) of them. Clinical diagnoses were corrected by autopsy findings when necessary. During the follow-up period, CVD (stroke in 155 and CHD in 34) occurred in 179 subjects who were not taking antihypertensive medication.

STATISTICAL METHODS

We calculated the incidences of CVD and its subtypes by the pooling of repeated observations method.[11] This technique is a generalized person-year approach that incorporates all repeated observations. By treating each examination interval as a miniature follow-up study, the method pools observations from all intervals to examine the short-term development of CVD. The incidences were compared, and relative risks were estimated by the time-dependent Cox proportional hazards model, in which risk factors were allowed to change in accordance with data from the 5 follow-up examinations.[12] If subjects had missing data in the pooling of repeated observations or in the time-dependent covariate model, they were excluded from these analyses. $P<.05$ was considered statistically significant. Statistical analyses were performed with the SAS program package (SAS Institute Inc, Cary, NC).

RESULTS

Trends in characteristics of subjects who had not been taking antihypertensive medication are shown in **Table 1**. Mean age increased from 68.7 years in 1961 to 88.2 years in 1988. The frequency of male subjects declined over time. The prevalence of hypertension (stages 1-3) slightly increased from 59.9% in 1961 to 69.2% in 1988. Body mass index slightly decreased from 21.2 in 1961 to 20.4 in 1988, whereas the mean total cholesterol level increased from 159 mg/dL (4.1 mmol/L) in 1961 to 193 mg/dL (5.0 mmol/L) in 1988. The frequency of smoking habits and alcohol intake declined with advancing age. The prevalence of glucose intolerance increased from 10.2% in 1961 to 20.0% in 1983, whereas in 1988, only 3.8% of the subjects were found to be glucose intolerant. Clear trends were not found in the frequency of abnormal ECG findings or proteinuria.

Table 2 shows the age-adjusted incidences and the multivariate-adjusted hazard ratios (HRs) of CVD among BP categories by sex. In men and women, the incidences and HRs of CVD increased with rising BP levels.

Table 1. Trends in Characteristics of Subjects*

Characteristic	Follow-up Years					
	1961 (n = 566)	1967 (n = 309)	1974 (n = 175)	1978 (n = 119)	1983 (n = 60)	1988 (n = 26)
Age, mean ± SD	68.7 ± 7.1	73.2 ± 6.0	79.3 ± 5.2	81.5 ± 4.5	85.6 ± 4.0	88.2 ± 2.2
Male, %	41.0	39.5	31.4	28.6	35.0	19.2
Blood pressure category, %†						
Optimal	15.2	15.9	9.7	12.6	10.0	7.7
Normal	11.5	13.6	9.7	5.9	5.0	19.2
High-normal	13.6	11.3	9.7	14.3	15.0	3.8
Stage 1	27.2	28.8	32.0	33.6	26.7	26.9
Stage 2	18.6	18.8	19.4	25.2	28.3	26.9
Stage 3	14.1	11.7	19.4	8.4	15.0	15.4
BMI, mean ± SD	21.2 ± 2.8	21.1 ± 2.9	21.3 ± 2.8	20.6 ± 3.2	20.4 ± 3.0	20.4 ± 3.9
Total cholesterol, mg/dL	159 ± 40	166 ± 40	167 ± 37	183 ± 37	191 ± 40	193 ± 36
Habitual smoking, %	38.7	35.3	25.7	20.2	21.7	15.4
Alcohol intake, %	30.9	22.3	21.7	20.2	15.0	0.0
Glucose intolerance, %	10.2	10.7	15.4	13.4	20.0	3.8
ECG abnormalities, %‡	21.6	22.1	25.1	21.0	31.7	19.2
Proteinuria, %	13.4	7.1	16.0	6.7	18.3	15.4

Abbreviations: BMI, body mass index (calculated as weight in kilograms divided by the square of height in meters); ECG, electrocardiography.
SI conversion factors: To convert total cholesterol to millimoles per liter, multiply by 0.0259.
*Unless otherwise indicated, data are expressed as percentages (frequencies).
†Categories are described in the "BP Classification and Risk Stratification" subsection of the "Methods" section.
‡ Indicates Minnesota codes 3-1 (high R-waves), 4-1, 4-2, and/or 4-3 (ST-segment depression).

Table 2. Age-Adjusted Incidences and Multivariate-Adjusted HRs of Cardiovascular Disease According to BP Category by Sex*

BP Category†	Men		Women	
	Incidence	HR (95% CI)	Incidence	HR (95% CI)
Optimal	9.7	1.00	1.6	1.00
Normal	21.5	1.32 (0.44-3.92)	0.1	0.42 (0.04-4.62)
High-normal	9.7	0.64 (0.19-2.15)	15.3	3.92 (0.84-18.27)
Stage 1	32.4	1.60 (0.68-3.75)	17.9‡	4.67 (1.10-19.84)§
Stage 2	32.5§	1.68 (0.69-4.09)	13.6§	3.36 (0.76-14.83)
Stage 3	75.8‡	2.72 (1.16-6.35)§	37.0‡	9.41 (2.16-46.97)‡

Abbreviations: BP, blood pressure; CI, confidence interval; HR, hazard ratio.
*Incidence rates are 1000 person-years. HRs are adjusted for age, body mass index, serum cholesterol level, smoking habits, alcohol intake, glucose intolerance, electrocardiographic abnormalities, and proteinuria.
†Categories are described in the "BP Classification and Risk Stratification" subsection of the "Methods" section.
‡$P<.01$ vs optimal BP.
§$P<.05$ vs optimal BP.

The age- and sex-adjusted incidences and the multivariate-adjusted HRs of CVD among BP categories are shown in **Table 3**. The incidences and HRs of stroke and CHD increased with elevating BP levels. When stroke and CHD were combined as total CVD, a similar association was observed. The differences between optimal BP and stages 1 through 3 hypertension were statistically significant even after adjustment for other covariates. In the following stratified analysis, the subjects in the optimal and normal BP groups were combined and used as a reference group, because the number of subjects categorized in each group was relatively small.

To examine whether the BP-CVD relationship changes with advancing age, the sex-adjusted incidences of CVD among BP categories were compared in 3 age strata (60-69, 70-79, and ≥80 years; **Figure 1**). Among subjects aged 60 to 79 years, the incidences increased with rising BP levels; the differences between optimal plus normal BP and stages 1 through 3 hypertension were statistically significant. In contrast, among participants 80 years or older, subjects with stage 3 hypertension alone had significantly higher incidences than did those with optimal or normal BP. Our findings demonstrated that the BP-CVD relationships in subjects no older than 79 years were different from those in very old subjects.

We further estimated the sex-adjusted incidences of CVD according to the risk stratification system in subjects aged 60 to 79 years and in those 80 years or older (**Figure 2**). In the younger subjects, as expected, the incidences increased with rising BP levels in each risk group. The incidences were significantly higher among subjects who had TOD or glucose intolerance (risk group C) compared with those of their counterparts (risk group B; sex- and BP-adjusted HR, 1.64; 95% confidence interval, 1.13-2.38; $P=.009$). In the oldest subjects in risk group B, the incidences of CVD were higher in patients with

Table 3. Age- and Sex-Adjusted Incidences and Multivariate-Adjusted HRs of Cardiovascular Disease by Type According to BP Category*

	Stroke		CHD		CVD	
BP Category†	Incidence	HR (95% CI)	Incidence	HR (95% CI)	Incidence	HR (95% CI)
Optimal	7.3	1.00	1.6	1.00	7.4	1.00
Normal	8.9	0.48 (0.15-1.51)	2.6	3.41 (0.35-33.20)	11.3	0.86 (0.32-2.27)
High-normal	12.5	1.00 (0.42-2.37)	2.9	3.32 (0.35-32.00)	15.2	1.43 (0.62-3.27)
Stage 1	23.8‡	1.92 (0.98-3.78)	5.5	2.96 (0.31-28.59)	27.9§	2.57 (1.30-5.12)§
Stage 2	23.8‡	1.76 (0.87-3.56)	5.5	3.30 (0.39-27.85)	25.5‡	2.36 (1.15-4.85)‡
Stage 3	61.7§	3.90 (1.96-7.75)§	5.7‡	5.28 (0.62-44.97)	65.0§	5.34 (2.66-10.71)§

Abbreviations: BP, blood pressure; CHD, coronary heart disease; CI, confidence interval; CVD, cardiovascular disease; HR, hazard ratio.
*Incidence rates are per 1000 person-years. HRs are adjusted for age, body mass index, serum cholesterol level, smoking habits, alcohol intake, glucose intolerance, electrocardiographic abnormalities, and proteinuria.
†Categories are described in the "BP Classification and Risk Stratification" subsection of the "Methods" section.
‡$P<.05$ vs optimal BP.
§$P<.01$ vs optimal BP.

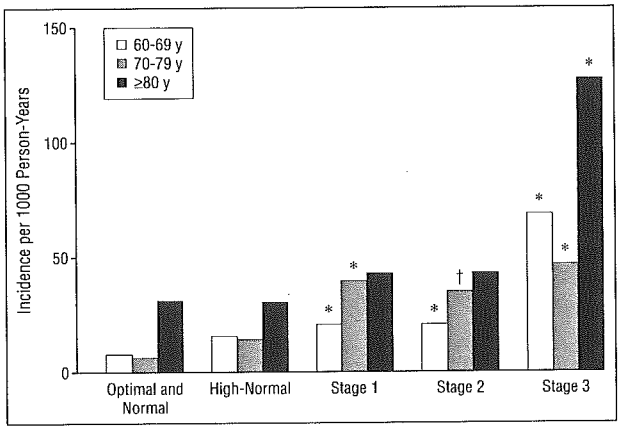

Figure 1. Sex-adjusted incidences of cardiovascular disease per 1000 person-years according to blood pressure category and 3 age strata. Asterisk indicates $P<.01$ vs optimal and normal blood pressure; dagger, $P<.05$ vs optimal and normal blood pressure.

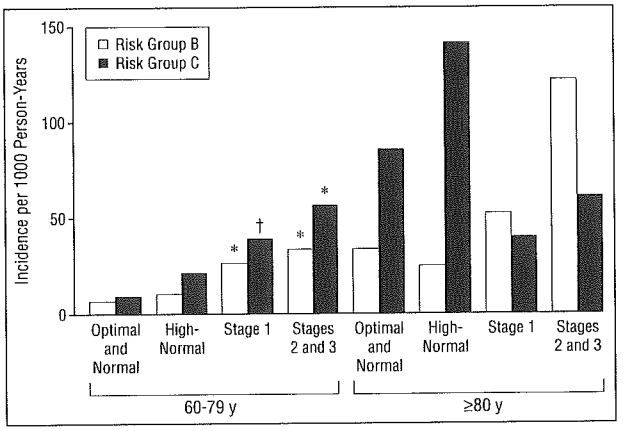

Figure 2. Sex-adjusted incidences of cardiovascular disease per 1000 person-years according to the risk stratification system in subjects aged 60 to 79 years and in those 80 years or older. Risk groups are described in the "BP Classification and Risk Stratification" subsection of the "Methods" section. Asterisk indicates $P<.01$ vs optimal and normal blood pressure; dagger, $P<.05$ vs optimal and normal blood pressure.

stages 1 through 3 hypertension than in those with optimal, normal, or high-normal BP, although these differences were not statistically significant. In contrast, no clear association was observed between BP and CVD among the oldest subjects with TOD or glucose intolerance (risk group C); the incidences of stages 1 through 3 hypertension tended to decrease.

COMMENT

We were able to observe the natural course of untreated hypertension during a 32-year period in a general population of Japanese elderly. An additional strength is that the transition of risk factors could be taken into account in the analysis by using the pooling of repeated observations method and time-dependent Cox proportional hazards model. In this study, we found a close association between BP and CVD incidence in the Japanese elderly. In addition, findings derived from subjects no older than 79 years support the JNC VI recommendations[1] to evaluate hypertension based on the BP level and the presence or absence of TOD or other risk factors. Our results indicate that treating hypertension according to the JNC VI risk stratification system, found useful in the US population,[2] might be useful in the Japanese elderly population.

Mortality due to stroke and CHD are well known to be heterogeneous among populations in different countries. Genetic heterogeneity and differences in risk factors may explain the differences in the stroke- and CHD-associated mortality. In the Seven Countries Study, the mortality due to stroke in Japan was 3 times higher compared with that in the United States,[13] and conversely, the mortality due to CHD in Japan was one third of that in the United States at the same BP level.[14] In other words, the mortality due to stroke was 5 times higher than that due to CHD in Japan, whereas the mortality due to CHD was twice as high as that due to stroke in the United States. However, the relative risk for death due to stroke and CHD as a function of BP elevation was similar between these 2 populations.[13,14] These facts may indicate that our results from Japanese elderly subjects are applicable to the US population.

The JNC VI treatment recommendations for older patients were the same as those for younger patients.[1] Japanese guidelines, in contrast, proposed initiating antihypertensive treatment at higher BP levels and also set

higher-goal BP levels among patients 70 years or older than those set for younger patients.[3] These recommendations were proposed on the basis of the results of several prospective studies.[15-17] In the Helsinki Aging Study,[15] BP was inversely correlated with mortality among individuals 75 years or older. Takagi et al[16] evaluated the long-term prognosis of CVD in Japanese elderly subjects aged 60 to 64 years and found that CVD mortality was significantly increased among patients with systolic BP of 160 mm Hg or higher. Port et al[17] investigated the relationship of mortality to systolic BP in the subjects in the Framingham Study by using the reduced horizontal–logistic spline model. They set a threshold at the 70th percentile of BP and concluded that the threshold increased with age. These observations may support the idea that BP levels at which to initiate antihypertensive treatment should be higher in the elderly. There are, however, several potential limitations to the findings in these studies. First, the relation between BP and events might be biased by inclusion of treated hypertensive patients. Second, hypertensive risk for CVD could be underestimated, because the outcomes of interest in these studies were total or CVD mortality, and nonfatal CVD events were not evaluated.

In the present study, we examined the contribution of hypertension to risk for CVD occurrence in Japanese elderly subjects not taking antihypertensive medication and found that among subjects no older than 79 years, the incidences were significantly higher in patients with stages 1 through 3 hypertension than in those with optimal and normal BP. In the Rotterdam Study, van den Hoogen et al[18] examined the association of BP with the risk for myocardial infarction among subjects not using BP-lowering medication and performed subgroup analyses for subjects aged 55 to 69 and 70 to 99 years. As a result, they found that a diastolic BP of 80 mm Hg or higher increased the risk for myocardial infarction in both age groups. In some other randomized trials, the benefits of antihypertensive treatment also persisted up to 80 years of age.[19-21] These data support the JNC VI recommendations[1] to administer the same treatment to older and younger patients with hypertension.

Previous studies have demonstrated that hypertensive risk for CVD was reduced with advancing age.[22,23] In subjects older than 80 years, the influence of BP on the development of CVD has been unclear. In several studies,[22,24,25] no clear association was found between BP and CVD mortality. In contrast, a report based on the Framingham Heart Study[26] documented significantly positive associations of BP with CVD incidence among subjects aged 75 to 94 years. A subgroup meta-analysis of randomized trials of participants 80 years or older demonstrated that antihypertensive treatment prevented 22% of major cardiovascular events.[27] In the present study, very old subjects with stage 3 hypertension alone had significantly higher incidences of CVD than subjects with optimal or normal BP. These data suggest that among subjects older than 80 years, very high BP may be a risk factor for CVD.

To our knowledge, the JNC VI risk stratification system[1] has not been evaluated in very old people. The findings derived from our subjects 80 years or older did not necessarily support this risk stratification system. In these oldest subjects without TOD and glucose intolerance, the incidences of CVD tended to increase with rising BP levels. However, no clear association between CVD incidences and BP levels was found in these oldest subjects with TOD or glucose intolerance. These findings cannot be considered incidental, because subjects 80 years or older contributed 26.8% of the total person-years of observation. One possible explanation is that in the oldest subjects with TOD or glucose intolerance, higher mortality due to other diseases (eg, heart failure, end-stage renal disease, or peripheral arterial disease, which is closely related to hypertension) may reduce the BP-CVD relationship. In our oldest subjects in risk group C, however, no positive association was observed between BP and non-CVD mortality (data not shown). Another explanation is that, as a result of more advanced coronary and cerebral atherosclerosis, high BP may be necessary to guarantee adequate blood flow in the myocardium and the brain in the oldest subjects with TOD or glucose intolerance. Kannel et al[26] demonstrated that the BP-CVD mortality associations were different between the older elderly subjects with and those without CVD. These data suggest that hypertension may not be a risk factor for CVD among the oldest subjects with advanced atherosclerosis.

Several potential limitations to the findings in our study exist. The primary limitation is that the definition of BP level was based on 3 measurements of BP taken on a single day. The BP levels might have been misclassified, despite the fact that measurements of BP on a single day have been suggested to be accurate in epidemiological studies.[28] Second, we excluded hypertensive patients who had started antihypertensive therapy, whose BP levels may have been higher. Given that these limitations might have reduced the estimate of the risk associated with high BP, the true association may be stronger than that suggested by our findings. Third, the risk stratification system we used in the present study was almost the same as, but not identical to, that of the JNC VI.[1] However, this bias did not seem to hold the potential to alter our findings' support of the general idea of the JNC VI treatment recommendations.

The present population-based study indicated that the management of hypertension according to the JNC VI recommendations[1] is applicable to elderly Japanese subjects up to 79 years of age. However, the beneficial effect of antihypertensive therapy for hypertensive subjects of this age, especially those with stage 1 hypertension, should be further clarified. In subjects 80 years or older with advanced atherosclerosis, on the other hand, we found no clear association of BP with CVD, which argues against the definite benefits of treating the oldest elderly patients with hypertension.

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REGULAR PAPER

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Age-associated prevalence and risk factors of Lewy body pathology in a general population: the Hisayama study

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Abstract In dementia with Lewy bodies (DLB), the Lewy bodies (LBs) are an essential substrate. Although LB pathology has gained increasing attention as one of the major causes of dementia, little is known about the exact prevalence of LB pathology in the general population. In addition, the pathology of Alzheimer-type dementia (ATD) is frequently associated with DLB. To investigate the prevalence of LB pathology in a community-based population and to evaluate the relationship between LB and ATD pathology, we performed an analysis of 102 consecutive autopsy cases. The survey extended over 2.5 years and autopsy rate was 70.5%. LB pathology was detected using α-synuclein immunohistochemistry and was assessed based on consensus guidelines for DLB. ATD pathology was evaluated by both CERAD and NIA-RI criteria. Twenty-nine subjects were clinically demented. LB pathology was present in 23 (22.5%) of 102 cases, and in 12 (41.4%) of the demented subjects. The LB score was not significantly different between DLB cases and non-demented subjects with LB pathology (nd-LB), while the Braak stages were significantly different between the two groups. Prevalence of LB pathology constantly increased with age. DLB cases accompanying severe ATD pathology showed more rapid increase of LB scores than did DLB cases without severe ATD pathology. Moreover, DLB cases with severe ATD pathology had poorer prognoses than those without severe ATD pathology. Our results suggested that aging and severe ATD pathology have a strong effect on the evolution of LB pathology.

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Keywords Lewy bodies · Dementia with Lewy bodies · Alzheimer-type dementia · Aging

Introduction

Recently, Lewy bodies (LBs) have been demonstrated to be primarily composed of aggregated α-synuclein, and well-characterized monoclonal antibodies that are sensitive and specific for α-synuclein have become available for analysis of paraffin-embedded sections [4, 17, 37]. With the development of this immunohistochemical staining, there is an increasing recognition of cortical LBs in demented patients, and LBs are considered an essential substrate for dementia with Lewy bodies (DLB). DLB is now the second most common form of dementia in the elderly, following Alzheimer-type dementia (ATD) [15, 16, 22, 24]. However, LB pathology is often found in a normal elderly population [8, 15, 32, 34]. Despite the marked impact of LB formation on the cause of dementia, little is known about the exact prevalence of LB pathology worldwide. The reported prevalences of LB pathology have varied from 2.3% to 60.7%, being mainly dependent on the sample selection and detecting method for LBs [8, 10, 12, 15, 23, 25, 30, 32, 34]. Because many previous reports have been based on the results of hospital-based postmortem studies and not on community-based studies, many subjects would have been collected with a bias towards clinically atypical cases. Furthermore, ATD pathology is frequently associated with DLB, but the precise nosological relationship between DLB and ATD remains an unresolved issue [9, 16, 26].

Since 1961, we have carried out a prospective population-based study in a Japanese subrural community, Hisayama Town, which is adjacent to the metropolitan area of Fukuoka on Kyushu Island, Japan. This study has investigated the epidemiology of cerebrovascular disease in a general Japanese population [19, 38]. A characteristic of the town is that the age, occupational status and nutrient intake of the population are almost identical to those of the general Japanese population [21, 41]. Moreover, the

population of the town is approximately 7,000 and has scarcely changed during the past 40 years. We carried out autopsies on most deceased subjects to confirm causes of death and to examine brain pathology. These features have allowed a reliable estimation of the frequency of LB pathology in a general population and for a detailed analysis of the relationship between DLB and ATD pathology.

To evaluate the accurate prevalence of LB pathology in a community-based population, we retrospectively analyzed 102 consecutive autopsy cases derived from Hisayama Town using α-synuclein immunohistochemistry (IHC). We also evaluated the influence of ATD pathology on DLB patients to elucidate the relationship between LB and ATD pathologies. In this study, we demonstrated that aging is one of the risk factors causing the prevalence of LB pathology. Moreover, we showed that severe ATD pathology might lead to the progression of LB pathology through the brain stem to the neocortex, worsening prognoses of DLB patients.

Materials and methods

Subjects

The prospective population survey on which the present study is based has been conducted in Hisayama since 1961 and is ongoing. We collected information about new neurological events, including stroke and cognitive impairment, through a daily monitoring system established by the study team, local practitioners, and the town government [41]. Members of our study group visited the town at least once a week to maintain contact with physicians and the staff of the local Health and Welfare Office. At least once a week, we also surveyed the three major hospitals with geriatric or psychiatric wards near the town, to which Hisayama residents are usually admitted when necessary. Regular health checks and extensive neuropsychiatric evaluation, including medical history and physical examination, neurological history and examination, semi-structured psychiatric interview, and neuropsychological assessment, were given biennially to obtain information on any new neurological events missed by the monitoring network. When we suspected new neurological symptoms, including cognitive impairment, the study physicians carefully evaluated the subject, and an effort was made to obtain further diagnostic information, including brain CT and MRI. The diagnosis of dementia was made clinically based on the guidelines of the Diagnostic and Statistical Manual of Mental Disorders, Revised Third Edition (DSM-III-R) [1]. For the clinical diagnosis of DLB, we used the consensus guidelines presented at the consortium on DLB (CDLB) International Workshop [26]. Vascular dementia (VD) and ATD were also clinically diagnosed by the criteria established by the National Institute of Neurological Disorders and Stroke-Association Internationale pour la Recherche et l'Enseignement en Neurosciences (NINDA-AIREN) International Workshop [36] and the National Institute of Neurological and Communicative Disorders and Stroke and the Alzheimer's Disease and Related Disorders Association (NINCDS-ADRDA) [28], respectively. The date of onset of VD was determined as the date when the responsible stroke occurred, but a final diagnosis of VD was made more than 3 months after the stroke. Because it is difficult to determine the onset of ATD, DLB, and VD when there is no antecedent stroke episode, a tentative time of onset, when the family or attending physician first noticed abnormal behavior by the subject, was used.

From 1 October 1998 to 31 March 2001, 149 Hisayama residents of all ages died; 105 of whom (70.5%) underwent postmortem examinations. Consent to autopsy was unobtainable from 28.5% of the residents due to refusal mainly on religious grounds. Of those 105 cases, 103 subjects received autopsies at the Departments of Pathophysiological and Experimental Pathology, Anatomic Pathology, and Neuropathology of Kyushu University. Autopsies of the 2 remaining cases were carried out at other institutes. Of the 103, 1 case showed respirator brain damage after subarachniod hemorrhage and we could not carry out a detailed histopathological examination, thus 102 cases were analyzed in this study.

Neuropathological assessment

Brains were weighed, evaluated for grossly detectable lesions and abnormalities of the blood vessels, and were fixed with 10% buffered formalin for at least 2 weeks. Brain specimens were taken following the Consortium to Establish a Registry for Alzheimer's Disease (CERAD) guidelines and the consensus guidelines for DLB [26, 27, 29]. Thus, the specimens in each case included middle frontal gyrus, superior and middle temporal gyri, inferior parietal lobule, anterior cingulate gyrus, hippocampus with entorhinal cortex and transentorhinal cortex (at the level of the lateral geniculate body, LGB), calcarine cortex, basal ganglia, thalamus, substantia nigra (SN), locus coeruleus (LC), and dorsal vagal nucleus (DVN). Sections were embedded in paraffin and were routinely stained using hematoxylin-eosin (HE) and a modified Bielschowsky's method.

Immunohistochemistry

IHC was performed on 7-μm paraffin-embedded sections. Sections were deparaffinized in xylene, hydrated in ethanol, and incubated with 0.3% hydrogen peroxide in absolute methanol for 30 min at room temperature to inhibit endogenous peroxidase. After rinsing with tap water, to enhance the immunoreactivity, the sections for α-synuclein and ubiquitin antibodies were completely immersed in 0.01 M citrate buffer, pH 6.0 and autoclaved at 121°C for 10 min. This antigen retrieval protocol for α-synuclein antibody, which had been previously reported elsewhere [20, 40], was as effective as formic acid or proteinase K pretreatment. After washing with TRIS-HCl buffer (50 mM TRIS-HCl, pH 7.6), mouse monoclonal antibody against α-synuclein (clone LB509, Dr. Iwatsubo) [4] diluted 1:100, and rabbit polyclonal antibodies against ubiquitin (Dako, Copenhagen, Denmark) diluted 1:1,000 and tau (Dako) diluted 1:200 were applied. The slides were incubated overnight at 4°C. Sections with α-synuclein antibody were then sequentially incubated for 1 h with biotinylated secondary antibodies diluted 1:200, and peroxidase conjugated streptavidin-biotin complex diluted 1:300 (Amersham, UK). Specimens with ubiquitin and tau antibodies were incubated for 1 h with horseradish peroxidase-conjugated goat anti-rabbit antibodies (Vector Laboratories, Burlingame CA) diluted 1:200. The reaction product was developed with 3,3'-diaminobenzidine tetrahydrochloride solution. The sections were lightly counterstained with hematoxylin.

Semiquantitative assessment of LB pathology

The extent of LB pathology was estimated using a scoring system based on that proposed in the consensus guidelines for DLB [26] with two modifications: (1) α-synuclein IHC was used additionally with ubiquitin IHC and HE staining to identify LB, and (2) the transentorhinal LB score was taken at the level of the LGB, which is slightly posterior to the section at the level of the red nucleus. In this modified system, specified portions of five areas of the brain: the transentorhinal cortex, cingulate gyrus, middle frontal gyrus, inferior parietal lobule, and middle temporal gyrus were examined. If there were more than five LBs, the area was given a score of 2. If there were one to five LBs, the score was 1. The scores for the five areas were then summed to yield the case's LB score. Cases were categorized into three groups: neocortical type (score 7–10), limbic type (score 3–6), and brain stem type (score 0–2). DLB cases were further divided into the "DLB/ATD" group and "pure DLB" group. The DLB/ATD group was defined as having severe

ATD pathology as well as LB pathology, whereas the pure DLB group was defined as being free from severe ATD changes.

Semiquantitative assessment of ATD pathology

Neuritic plaques were estimated by a modified Bielschowsky's method. Neurofibrillary tangles (NFT) were assessed by tau IHC. In each case, the frequency of neuritic plaques and NFT were semiquantitatively evaluated, and converted to a plaque score according to CERAD criteria and Braak stage established by Braak and Braak [5, 29]. The CERAD score and the Braak stage were combined to estimate the likelihood that dementia was due to ATD according to the NIA-RI criteria [2]. A diagnosis of ATD or severe ATD pathology was made when "definite ATD" as defined by the CERAD criteria and/or a "high-likelihood" as defined by the NIA-RI criteria were determined.

Assessment of VD pathology

For the pathological assessment of vascular dementia, we used criteria presented at the NINDA-AIREN International Workshop [36]. All infarcts (including status lacunaris and Binswanger's disease/leukoaraiosis) and hypertensive hemorrhages were registered with regard to their age, size, and topographical location.

Assessment of MD pathology

Although the term 'mixed dementia (MD)', referring to the coexistence of clinicopathological features of ATD and VD, has been used worldwide, MD remains an unclear diagnosis. We tentatively diagnosed MD as follows: the presence of both VD pathology corresponding to NINDA-AIREN criteria and severe ATD pathology described above. If severe ATD pathology was thought to be responsible for dementia, and the associated foci of cerebrovascular lesions alone were likely not to be the cause of dementia, the case was classified as ATD.

Statistical analysis

Statistical analysis was performed using StatView program J-5.0 for Macintosh. The differences between two groups were analyzed using the Student's *t*-test. Non-parametric Mann-Whitney U-test was used to analyze the differences in LB score and Braak stage between DLB subjects and non-demented subjects with LB pathology (nd-LB), between pure DLB and DLB/ATD, and between DLB and ATD cases. Spearman's rank correlation test was carried out to evaluate the correlation between the Braak stage and LB score among the LB-positive subjects, and the correlation between the duration of dementia and LB score among the DLB cases. Cumulative survival rate was evaluated using a Kaplan-Meier test and the difference between two survival curves was analyzed by log rank test.

Results

Clinico-neuropathological information of all subjects

The mean age at death of the 102 consecutive autopsies was 80.2±12.2 years, and gender was evenly distributed (51 males/51 females) (Table 1). Twenty-nine cases (28.4%) presented clinical signs of dementia. The age at death in demented patients was significantly higher than that in non-demented individuals (*t*-test, $P<0.01$). Among the demented cases, females dominated (20 females/9 males), whereas among the non-demented subjects the distribution of gender tended to be opposite (31 females/42 males). Brain weight was significantly lower in the demented than in non-demented subjects (*t*-test, $P<0.01$). LB pathology was present in 23 (22.5%) of the 102 cases. Among the non-demented group ($n=73$), 11 cases (15.1%) showed LB formation. None of these nd-LB cases were clinically diagnosed with Parkinson's disease (PD). Of the 29 demented subjects, 12 cases (41.4%) presented LB formations and had a pathological diagnosis of DLB. Four cases (13.8%) diagnosed as 'pure' ATD (ATD without LB pathology), 9 cases (31.0%) as VD, 3 cases (10.3%) as MD, and 1 case had another neuropathological diagnosis. The LB scores appeared to be greater in DLB cases than in nd-LB subjects, but the difference was not significant (Mann-Whitney U-test, $P=0.08$) (Table 2). Braak stages were significantly greater in the DLB cases versus nd-LB subjects (Mann-Whitney U-test, $P<0.01$) (Table 2). The neocortical type of DLB also showed significant higher Braak stages as compared with the neocortical type of nd-LB subjects (Mann-Whitney U-test, $P<0.05$) (Table 2). Although the Braak stage of every ATD cases were stage 6, and tended to show a higher Braak stage than that of DLB cases, the different was not significant (Mann-Whitney U-test, $P=$

Table 1 Clinical and pathological information of all subjects. Values for the last four columns are given as mean ± SD (*DLB* dementia with Lewy bodies, *ATD* Alzheimer-type dementia, *MD* mixed dementia, *VD* vascular dementia)

Group	*n* (male/female)	Age at onset (years)	Age at death (years)	Duration (years)	Brain weight (g)
Total	102 (51/51)		80.2±12.2		1,221.6±161.6
Non-demented subjects	73 (42/31)		77.4±12.6[a]		1,256.4±157.1[b]
with LB pathology	11 (5/6)		84.5±6.9		1,199.5±96.2
with ATD pathology	3 (1/2)		80.7±11.0		1,131.7±325.1
Demented patients	29 (9/20)	82.3±8.1	87.5±7.2[a]	5.3±4.1	1,134.0±139.7[b]
DLB	12 (3/9)	82.2±9.5	88.2±8.2	5.8±5.1	1,149.6±128.8
pure DLB	5 (1/4)	78.6±10.0	89.0±8.2	10.0±5.5[c,d]	1,082.0±157.7
DLB/ATD	7 (2/5)	84.7±9.0	87.6±8.7	2.9±1.6[c]	1,197.9±85.6
ATD	4 (1/3)	82.8±6.7	86.0±6.3	4.2±1.4	1,068.0±50.4
MD	3 (0/3)	89.3±8.0	95.0±6.9	5.4±3.1	1,120.0±156.0
VD	9 (5/4)	80.9±6.0	84.9±5.6	4.3±3.8[d]	1,182.5±113.6
Others	1 (0/1)	70	81	11.3	755

Significant difference between the two values [a]$P<0.01$, [b]$P<0.05$, [c]$P<0.01$, [d]$P<0.05$