Introduction

Antihypertensive therapy is essential to reduce the occurrence of cardiovascular (CV) events and mortality. Many large-scale studies of various antihypertensive medications, such as Ca channel blockers (CCBs), angiotensin receptor antagonists (ARBs) and diuretics, have shown that reduction of the blood pressure (BP) is essential to prevent CV events (1–4). Based on these results, guidelines for the clinical management of hypertension such as Japanese Society of Hypertension Guidelines for the Management of Hypertension (JSH 2004) (5) and the recommendations of the Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC 7) (6) have been established and are being used in the daily management of hypertension. According to these guidelines, the optimal target BP level for uncomplicated hypertension is defined as lower than 140/90 mmHg regardless of age, although an interim level of 150/90 mmHg for elderly patients aged 75 or older is recommended for the purpose of careful and gradual reduction to the optimal target level in the Japanese guidelines.

The optimal target BP levels based on the risk factors are also established in these guidelines, but these levels are mainly derived from studies performed in Western countries, which enrolled patients who met the various inclusion and exclusion criteria. In clinical practice, clinicians prescribe antihypertensive medications daily for patients with various background factors, such as age, complications, and history of CV events that differ from those of the subjects in controlled clinical trials. Therefore, it is very important to assess the influence of various backgrounds as risk factors on antihypertensive therapy and the risk of future CV events. Several studies of Japanese hypertensive patients have already been conducted, but these studies have mainly assessed small cohorts in specific rural areas. Information from large-scale studies under daily clinical practice is limited.

The Japan Hypertension Evaluation with Angiotensin II Antagonist Losartan Therapy (J-HEALTH) study was initiated in 2000 as a large-scale observational study of losartan therapy. This study was designed to enroll 30,000 patients with hypertension throughout Japan, and the subjects were treated with losartan on an open-label basis mainly at a daily dose of 50 mg under common clinical management for a maximum of 5 years (7). The present study focuses on the relation between BP control and the incidence of CV events in not only the overall patient group but also elderly or diabetic patients of the J-HEALTH cohort.

Methods

Subjects

The eligible patients were hypertensive men or women aged 20 years or older who had not taken any antihypertensive agents within the previous 1 month. Patients who had previously been treated with losartan were excluded. Each patient was informed of the purpose and methods of the study, as well as the measures taken for protection of privacy, before they were enrolled. Patients gave verbal informed consent and then underwent a complete review of their medical history, as well as physical examination and laboratory evaluation. The methods were previously reported in detail (7).

Treatment and Monitoring

The patients were initially treated with losartan at a dose of 25–50 mg once daily, which was increased up to 100 mg once daily if necessary. Addition of other antihypertensive agents was allowed from 3 months after the start of losartan therapy, if required. No restrictions were placed on the treatment of complications.

The clinic BP was measured by the usual method at each institution. At each time of measurement, one clinic BP value was reported at the discretion of the physician. After starting losartan therapy, the clinic BP value was measured every 3 months for analysis of the clinic BP values during treatment. Standard laboratory tests were performed every 6 months with the routine methods used at each institution. To assess complications and the medical history, physicians judged the existence of disease indicated in the registration form at their discretion.

In addition, the patients who were on drug treatment for hyperlipidemia or diabetes mellitus and met the definition of either disease indicated in the relevant guidelines were defined as having hyperlipidemia or diabetes. Hyperlipidemia also included at least one of the dyslipidemic constituents, such as total cholesterol (TC) ≥ 220 mg/dL, low-density lipoprotein-cholesterol (LDL-C) ≥ 140 mg/dL, high-density lipoprotein-cholesterol (HDL-C) < 40 mg/dL, and triglycerides (TG) ≥ 150 mg/dL. Diabetes mellitus was defined as a history of diabetes mellitus, or fasting blood glucose (FBG) > 126 mg/dL and/or HbA1c $> 6.5\%$.

Evaluation of Endpoints

The primary endpoint of the study was a composite of CV events including fatal or non-fatal stroke (new occurrence or recurrence of cerebral hemorrhage, cerebral infarction, or subarachnoid hemorrhage diagnosed on the basis of typical clinical symptoms persisting for more than 24 h and/or computerized tomography/magnetic resonance imaging findings), transient ischemic attack (TIA) defined as a focal neurological deficit presumed to be vascular in origin persisting for less than 24 h, fatal or non-fatal myocardial infarction (MI) (new occurrence or recurrence) diagnosed on the basis of typical clinical symptoms, electrocardiogram changes and elevation of cardiac enzymes, or sudden cardiac death. In addition, the independent event classification committee reviewed adjudi-

Table 1. Baseline Characteristics of the Patients

	Total (*n*=26,512)	Male (*n*=11,638)	Female (*n*=14,874)
Age (years)	62.2±12.0	59.9±12.0	64.0±11.8
Clinic SBP (mmHg)	165.8±17.1	165.0±16.8	166.5±17.2
Clinic DBP (mmHg)	94.8±11.5	96.7±11.4	93.3±11.4
BMI (kg/m^2)	24.1±3.5	24.3±3.3	23.9±3.7
Alcohol drinking (%)	38.8	68.1	16.0
Smoking habit (%)	25.1	44.9	9.7
Complications			
Hyperlipidemia (%)	38.3	36.0	40.2
Diabetes mellitus (%)	12.6	15.3	10.5
Hyperuricemia/Gout (%)	10.6	19.8	3.5
Cardio-/cerebrovascular disease (%)	10.5	10.9	10.2

Data are expressed as mean±SD. SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index. Alcohol drinking: ≥3 times/week and ≥200 mL/time (1 middle-size bottle of beer or 2 glasses of diluted whiskey with water).

cated endpoint events on the basis of all available information documented in the case report form by the physicians.

Statistical Analysis

For the present analysis, variables were compared using the *t*-test, the χ^2 test, or analysis of variance (ANOVA). Results were expressed as the mean±SD, and differences were considered statistically significant at $p<0.05$. Analysis of the overall results was based on survival analysis. Subgroups were stratified by BP values measured during treatment. Relationships between the endpoints and BP values or prognostic factors were assessed by using the Cox proportional hazards model. Statistical analyses were conducted with the SAS package (Version 8.02; SAS Institute Inc., Cary, USA).

Results

Subjects and Follow-Up

Out of 31,048 patients enrolled in the study, 4,536 patients were excluded because of protocol violations or lack of data, and thus a total of 26,512 patients were used to investigate the relationship between BP and stroke or MI. The mean follow-up period was 3.0 years. The clinical characteristics of the 31,048 enrolled patients at baseline have already been reported (7), and the characteristics of the 26,512 patients analyzed in the present study are shown in Table 1. The profile of these patients was similar to that of the total group of all enrolled patients. Their mean age was 62.2±12.0 years and the mean baseline BP was 165.8±17.1/94.8±11.5 mmHg. Major complications were hyperlipidemia (38.3%) and diabetes (12.6%). The concomitant antihypertensive medications have been described elsewhere (8). In brief, 41% of the subjects were taking two or more antihypertensive medications. Their BP decreased to 136.9±13.2/79.2±9.6 mmHg after 60 months.

Table 2. Incidence of Stroke and MI during Treatment

Events	No. of events	Incidence
Stroke	307	3.90
Cerebral infarction	205	
Transient ischemic attack	19	
Cerebral hemorrhage	55	
Subarachnoid hemorrhage	20	
Unclassified stroke	8	
MI	80	1.02
Total	387	4.92

No. of patients: 26,512. Incidence: events/1,000 patient-years. MI, myocardial infarction including sudden cardiac death.

Incidence of Stroke and MI, and Risk Factors for Stroke and MI

During the follow-up period, cerebrovascular events (stroke or TIA) and MI occurred in 307 and 80 patients, respectively. The incidences of stroke or TIA and MI during treatment were 3.90 and 1.02 per 1,000 patient-years, respectively (Table 2). The incidence of stroke was 4-fold higher than that of MI.

Next, we identified risk factors contributing to CV events by using the Cox proportional hazards model. The relative risk of CV events was highest at an age over 75 years (3.81), while current smoker status (1.88), CV disease (1.97), diabetes mellitus (1.51), and hyperuricemia (1.37) were also significant risk factors for the occurrence of CV events (Table 3).
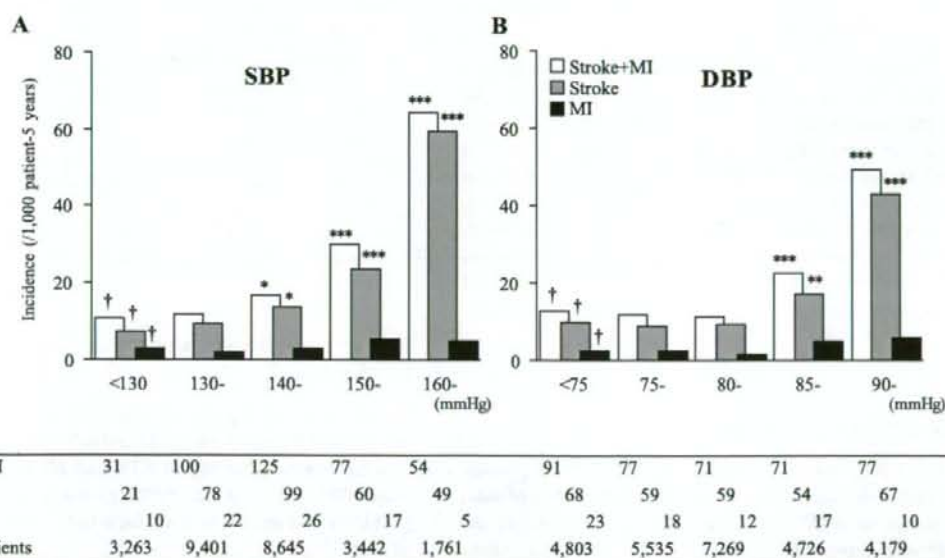
Relationship between BP and CV Events

The relationship between BP during treatment and the incidence of CV events is shown in Fig. 1. The incidence of CV events increased along with the BP and was significantly higher when the systolic BP (SBP) was higher than 140

Table 3. Relative Risk of Stroke and MI for Baseline Characteristics

	No. of patients	No. of events	RR (95% CI)	*p* value
Male	11,638	194	1.36 (1.07–1.73)	0.013
Age (years)				
<65	14,646	121	1.00 (Reference)	
65–74	7,690	131	1.93 (1.50–2.49)	<0.001
≥75	4,176	135	3.81 (2.92–4.97)	<0.001
Obesity (BMI ≥25 kg/m²)	7,452	105	1.06 (0.84–1.35)	0.621
Smoking habit	6,666	128	1.88 (1.45–2.45)	<0.001
Alcohol drinking	10,295	149	0.85 (0.65–1.12)	0.247
Cardio-/cerebrovascular disease	2,793	97	1.97 (1.55–2.50)	<0.001
Hyperlipidemia	10,163	163	1.10 (0.89–1.35)	0.375
Diabetes mellitus	3,345	76	1.51 (1.17–1.94)	0.001
Hyperuricemia	2,812	55	1.37 (1.02–1.84)	0.036
Urinary protein ≥ +	1,390	32	1.46 (1.00–2.13)	0.053

RR, relative risk; MI, myocardial infarction including sudden cardiac death; BMI, body mass index; CI, confidence interval. Alcohol drinking: ≥3 times/week and ≥200 mL/time (1 middle-size bottle of beer or 2 glasses of diluted whiskey with water). Adjusted for sex, age, diabetes mellitus, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking.

Stroke + MI	31	100	125	77	54	91	77	71	71	77
Stroke	21	78	99	60	49	68	59	59	54	67
MI	10	22	26	17	5	23	18	12	17	10
No. of patients	3,263	9,401	8,645	3,442	1,761	4,803	5,535	7,269	4,726	4,179

Fig. 1. *Relationship between the incidence of cardiovascular events and (A) SBP or (B) DBP level during antihypertensive treatment. SBP, systolic blood pressure; DBP, diastolic blood pressure; MI, myocardial infarction including sudden cardiac death. The results were adjusted for sex, age, diabetes mellitus, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking. *p<0.05, **p<0.01, ***p<0.001 vs. †reference category.*

mmHg and the diastolic BP (DBP) was higher than 85 mmHg than when BP was lower than 130/75 mmHg. The relationship between MI and BP was not clearly shown because of the low incidence of MI in the J-HEALTH cohort (Table 2). The incidence of stroke was strongly correlated with BP, and similar results were also observed when stroke was separated into cerebral hemorrhage and infarction (data not shown).

The relationship between BP and the incidence of CV events with and without various complications is shown in Table 4. In diabetic patients, the risk was increased at 130/85 mmHg or higher. In patients with a history of cerebrovascular or cardiovascular diseases, the incidence of stroke and MI

Table 4. Relationship between Blood Pressure during Treatment and Incidence of Stroke or MI in Patients with/without DM or CVD

	Non-DM				DM				Non-CVD				CVD			
	n	Events	Incidence	*p*	*n*	Events	Incidence	*p*	*n*	Events	Incidence	*p*	*n*	Events	Incidence	*p*
SBP (mmHg)																
<130	2,898	25	10.0	†	365	6	18.2	0.189	2,787	20	10.1	†	476	11	22.2	0.034
130–139	8,302	79	11.1	0.659	1,099	21	19.3	0.027	8,399	68	10.4	0.910	1,002	32	28.0	<0.001
140–149	7,559	99	15.9	0.04	1,086	26	25.3	<0.001	7,817	97	16.0	0.059	828	28	31.3	<0.001
150–159	2,923	60	28.5	<0.001	519	17	40.5	<0.001	3,110	61	29.1	<0.001	332	16	48.8	<0.001
≥160	1,485	48	74.8	<0.001	276	6	35.5	0.005	1,606	44	61.1	<0.001	155	10	111.4	<0.001
DBP (mmHg)																
<75	4,035	71	12.6	†	768	20	17.7	0.173	3,955	60	12.4	†	848	31	23.0	0.005
75–79	4,800	63	11.4	0.564	735	14	16.4	0.357	4,879	59	11.7	0.769	656	18	18.7	0.121
80–84	6,387	56	10.1	0.231	882	15	18.7	0.163	6,606	55	10.5	0.375	663	16	20.1	0.085
85–90	4,230	56	20.5	0.008	496	15	39.9	<0.001	4,359	50	18.6	0.038	367	21	60.4	<0.001
≥90	3,715	65	49.2	<0.001	464	12	56.7	<0.001	3,920	66	47.1	<0.001	259	11	79.6	<0.001
Total	23,167	311	16.6		3,345	76	24.9		23,719	290	16.3		2,793	97	31.7	

Incidence: events/1,000 patient-years. MI, myocardial infarction including sudden cardiac death; DM, diabetes mellitus; CVD, cardio-/cerebrovascular disease; SBP, systolic blood pressure; DBP, diastolic blood pressure. Adjusted for sex, age, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking in non-DM and DM. Adjusted for sex, age, diabetes mellitus, smoking habit and alcohol drinking in non-CVD and CVD. †Reference category.

increased significantly at a much lower level of SBP than in those without a history of these diseases.

BP and CV Events in Elderly Patients

We also analyzed the relationship between BP during treatment and CV events in elderly patients. Patients were divided into three age groups, which were ≥75 years (oldest, *n*=4,176), 65 to 74 years (older, *n*=7,690), and <65 years (middle-aged, *n*=14,646) according to the age classification of the JSH guidelines. All CV events occurred in 121 patients of the middle-aged group, in 131 patients of the older group, and in 135 patients of the oldest group.

As shown in Fig. 2, the incidence of CV events was strongly related to BP in all three age groups. Compared with the DBP, control of the SBP had more influence on the increase of CV events. Figure 2 also demonstrates that the BP during treatment was more strongly related with the incidence of CV events in younger patients than in older patients. In patients under 65 years old, the incidence of CV events was 9-fold higher at an SBP >160 mmHg than at an SBP <130 mmHg. In contrast, the incidence was only 4 times higher in patients over 75 years old in the same comparison. However, the absolute incidence of CV events was much higher in the older age groups. The incidences of CV events per 1,000 patient-years during 5 years of follow-up at an SBP of less than 130 mmHg were 5.8 for the middle-aged (<65 years old), 14.4 for the older (≥65 to <75 years old) and 32.8 for the oldest group (≥75 years old).

A few clinical data in very elderly patients were available; however, we had 692 patients aged 85 or older in the present study. So we analyzed the incidence of CV events in this very elderly subset. BP was well-controlled below 140/90 mmHg in about half of these very elderly Japanese patients with hypertension, and the incidence of CV events was 2-fold higher in the group with a BP ≥140/90 mmHg (96.8 per 1,000 patient-years for 5 years of follow-up) than in those with a BP <140/90 mmHg (42.4 per 1,000 patient-years for 5 years of follow-up) (Fig. 3). As shown in Table 5, the baseline characteristics in these two groups were similar except for BP levels.

BP and Mortality

All-cause mortality was 5.67/1,000 patient-years (446/26,512), while cardiovascular morality (death from stroke, TIA, MI, or cardiac sudden death) was 0.8/1,000 patient-years (63/26,512). A J-shaped curve was observed between total mortality and SBP or DBP level. Cardiovascular mortality increased with an elevation of SBP, but a J-shaped relationship was observed between DBP and cardiovascular mortality (Table 6). Neither SBP nor DBP affected cancer mortality in this study (data not shown). Total mortality in patients with SBP <130 mmHg was significantly higher than that in patients with SBP 130–139 (*p*<0.05) and 140–149 (*p*<0.05) mmHg. No significant difference in total mortality was seen among patients with SBP <130, 150–159, and ≥160 mmHg. Cardiovascular mortality in patients with SBP ≥160 mmHg was 6-fold higher (*p*<0.001) than that in patients with SBP <130 mmHg. Total mortality was significantly lower in patients with DBP 75–79 mmHg (*p*<0.01), and significantly higher in patients with DBP ≥90 mmHg (*p*<0.01) than that in patients with DBP <75 mmHg. A similar pattern was
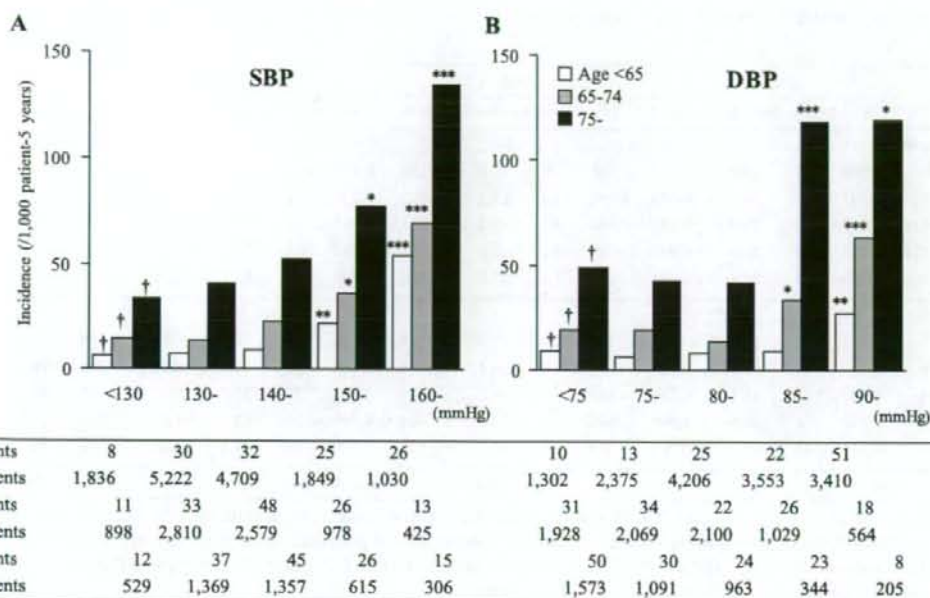
Fig. 2. *Relationship between the incidence of cardiovascular events and (A) SBP or (B) DBP level during antihypertensive treatment in 3 different age groups. SBP, systolic blood pressure; DBP, diastolic blood pressure; MI, myocardial infarction including sudden cardiac death. The results were adjusted for sex, diabetes mellitus, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking. *p < 0.05, **p < 0.01, ***p < 0.001 vs. †reference category.*

		<130	130-	140-	150-	160-		<75	75-	80-	85-	90-
<65	Events	8	30	32	25	26		10	13	25	22	51
	Patients	1,836	5,222	4,709	1,849	1,030		1,302	2,375	4,206	3,553	3,410
65-74	Events	11	33	48	26	13		31	34	22	26	18
	Patients	898	2,810	2,579	978	425		1,928	2,069	2,100	1,029	564
≥75	Events	12	37	45	26	15		50	30	24	23	8
	Patients	529	1,369	1,357	615	306		1,573	1,091	963	344	205

observed between cardiovascular mortality and DBP levels. Most notably, the mean age in patients with DBP <75 mmHg was approximately 15 years older than that in patients with DBP ≥90 mmHg (69.6 *vs.* 54.7 years old) (Table 6).

Discussion

The J-HEALTH study is a large-scale (30,000 patients) nationwide multicenter observational study that is providing valuable epidemiological information about Japanese patients with hypertension. We have previously reported on the clinical characteristics of the J-HEALTH cohort (7), which is a relatively young population with mild hypertension and few complications. Because our cohort was enrolled all over Japan (data not shown), the subjects are thought to be representative of the actual patients treated in daily Japanese clinical practice.

Staessen *et al.* performed a meta-analysis and concluded that lowering of the BP was needed for the prevention of CV events (4). Therefore, it is important to assess not only the BP at baseline, but also the mean reduction of BP during observation. Overall BP control in the J-HEALTH cohort ($n = 26,512$) has been described in detail elsewhere (8). We summarized the BP status at baseline and during observation for these patients in the present study. The baseline BP and the reduction of BP during losartan-based antihypertensive treatment

were 165.8/94.8 mmHg and 24.4/13.9 mmHg, respectively. Baseline BP and the reduction of BP during the Losartan Intervention For Endpoint reduction (LIFE) (2), the Antihypertensive and Lipid-Lowering Treatment to Prevent Heart Attack Trial (1), and the Controlled Onset Verapamil Intervention of Cardiovascular End Points studies (9) were, respectively, 174.3/97.9 and 30.2/16.8; 142/84 and 12.3/8.6; and 150.1/86.8 and 13.6/7.8 mmHg. Compared with these trials, our data suggest that the reduction of BP achieved by current clinical management in Japan is comparable with the results of other randomized controlled trials.

We investigated the incidence of CV events (stroke and MI). As shown in Table 2, CV events occurred in 387 patients (4.92/1,000 patient-years). Tanizaki *et al.* reported that the incidence of cerebral infarction was 6.4/1,000 patient-years for men and 3.4 for women in the Hisayama study (10). The incidence of stroke in the J-HEALTH study was similar to that in the Hisayama study. Regarding MI, its incidence in patients with hypercholesterolemia was 0.91/1,000 patient-years in the Japan Lipid Intervention Trial (J-LIT) study (11), and the incidence of MI was similar in the present study.

We also investigated the relative risk of CV events for each baseline characteristic. The results clearly demonstrated that well-known risk factors, such as age, smoking, a history of CV disease, diabetes mellitus, and hyperuricemia, were independent contributors to the development of CV events during
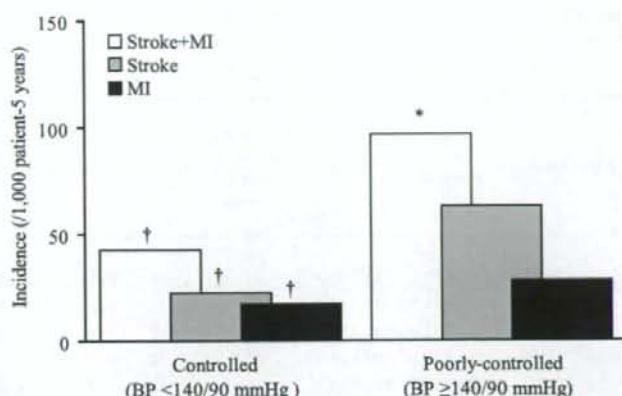
	Controlled	Poorly-controlled
Stroke + MI	9	18
Stroke	5	12
MI	4	6
No. of patients	310	382

Fig. 3. *Incidence of cardiovascular events in patients aged 85 years or older. BP, blood pressure; MI, myocardial infarction including sudden cardiac death. The results were adjusted for sex, diabetes mellitus, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking.* *$p < 0.05$ vs. †reference category.*

Table 5. Characteristics of Patients Aged 85 Years or Older

	Controlled BP <140/90 mmHg (n=310)	Poorly controlled BP≥140/90 mmHg (n=382)	p value
Baseline			
Male (%)	28.1	23.6	0.177
Age (years)	87.8±2.8	87.7±2.9	0.646
BMI (kg/m²)	21.6±3.6	21.5±3.2	0.743
Smoking habit (%)	11.3	8.4	0.226
Alcohol drinking (%)	12.5	12.1	0.882
SBP (mmHg)	162.9±17.0	172.1±17.8	<0.001
DBP (mmHg)	85.7±12.2	86.9±12.4	0.231
Cardio-/cerebrovascular disease (%)	40.3	23.3	<0.001
Diabetes mellitus (%)	13.6	8.6	0.039
Hyperlipidemia (%)	28.7	27.8	0.780
During treatment			
SBP (mmHg)	131.3±6.8	151.5±10.3	<0.001
DBP (mmHg)	72.8±6.6	78.9±8.5	<0.001
Concomitant antihypertensive drugs (%)	42.9	49.0	0.113

Data are expressed as mean±SD. BP, blood pressure; SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index. Alcohol drinking: ≥3 times/week and ≥200 mL/time (1 middle-size bottle of beer or 2 glasses of diluted whiskey with water).

daily clinical practice in Japan. We could not demonstrate that hyperlipidemia was a significant independent risk factor for CV events. Although the J-HEALTH cohort had a high percentage of hyperlipidemic patients (38.3%), their mean serum TC level was not particularly high, possibly because lipid levels in the majority of patients were relatively low. This could

be one of the reasons why we could not detect an influence of hyperlipidemia on CV events.

Although several interventional studies (*1, 2, 9*) have shown that BP control is beneficial for hypertensive patients, it is also valuable to demonstrate the influence of BP on CV events in actual clinical practice. The Hisayama study is a

Table 6. Incidence of Total and Cardiovascular Deaths in Patients Stratified with Blood Pressure during Treatment

	All patients (n=26,512)		Total death					Cardiovascular death (stroke and MI)				
	n	Age (years old)	Events	Incidence	Age (years old)	SBP/DBP (mmHg) (mean values during treatment)	PP (mmHg) (mean values during treatment)	Events	Incidence	Age (years old)	SBP/DBP (mmHg) (mean values during treatment)	PP (mmHg) (mean values during treatment)
SBP (mmHg)												
<130	3,263	61.9±12.4	67	45.4†	77.1±10.6	123.1/72.4	50.8	6	1.4†	71.3±11.0	126.0/76.7	49.3
130–139	9,401	62.0±11.8	143	33.7*	73.7±11.2	135.2/77.0	58.2	21	1.8	71.3±10.7	135.2/78.0	57.2
140–149	8,645	62.4±11.9	127	33.1*	74.1±11.0	144.4/78.0	66.4	12	1.2	77.1±8.5	145.7/77.8	67.9
150–159	3,442	62.6±12.4	85	61.0	75.1±12.3	154.3/81.6	72.7	14	3.8*	77.5±12.3	154.3/82.3	72.0
≥160	1,761	62.0±12.4	24	56.5	73.3±14.1	167.9/88.3	79.6	10	8.9***	67.4±15.7	167.6/89.7	77.9
DBP (mmHg)												
<75	4,803	69.6±10.5	160	38.1†	78.9±9.2	136.5/68.9	67.6	23	1.9†	77.0±8.9	140.3/69.9	70.4
75–79	5,535	65.4±11.0	92	26.0**	74.2±10.3	141.0/77.4	63.6	7	0.7*	76.0±9.6	143.8/78.4	65.4
80–84	7,269	61.8±11.4	100	31.1	73.4±10.9	141.7/82.2	59.5	8	0.9	73.3±13.1	149.0/83.4	65.6
85–90	4,726	58.3±11.0	58	42.0	72.0±13.4	146.8/86.9	59.9	14	3.5	73.9±13.2	147.0/86.9	60.1
≥90	4,179	54.7±11.1	36	62.8**	63.7±12.2	154.8/94.6	60.2	11	6.5**	62.5±11.6	154.2/94.8	59.4

Data are expressed as mean±SD. Incidence: events/1,000 patient-5 years. MI, myocardial infarction including sudden cardiac death; SBP, systolic blood pressure; DBP, diastolic blood pressure; PP, pulse pressure. Adjusted for sex, age, diabetes mellitus, cerebrovascular disease, cardiovascular disease, smoking habit and alcohol drinking. *$p<0.05$, **$p<0.01$, ***$p<0.001$ vs. †reference category.

long-term observational study of Japanese patients. It has demonstrated that a SBP >140 mmHg leads to a higher mortality rate compared with a BP ≤140 mmHg (*12*). Figure 1 indicates that there was a clear relation between BP during treatment and CV events, especially stroke, suggesting that the BP level should be less than 140/85 mmHg in Japanese with hypertension treated clinically. The lack of any association between BP and the incidence of MI may have been related to the low incidence of MI in our present population.

The target BP was examined in hypertensive patients with diabetes in the Hypertension Optimal Treatment study. The risk of CV events was significantly lower in patients with DBP ≤80 mmHg than in those with ≤85 mmHg or ≤90 mmHg (*13*). It was recently reported for Japanese patients that the risk of CV events was significantly decreased at BP ≤130/80 mmHg in hypertensive patients with diabetes in the J-LIT sub-analysis (*14*). As previously reported, the risk of CV events was lower at BP <130/85 mmHg in hypertensive patients with diabetes in the J-HEALTH cohort. These findings from J-HEALTH data support the JSH 2004 (*5*), the JNC 7 (*6*), the American Diabetes Association (*15*) and the European Society of Hypertension/European Society of Cardiovascular (ESH/ESC) (*16*) guidelines for treatment of hypertension accompanied with diabetes mellitus, which recommended that the target SBP/DBP be lower than 130/80 mmHg.

In the stratified analysis by age, the incidence of CV events in younger patients was more strongly influenced by BP elevation than that in elderly patients (Fig. 2). The Prospective

Studies Collaboration has published a meta-analysis of individual data on BP and mortality for one million adults taken from 61 prospective observational studies (*17*). Although they analyzed mortality, BP was linearly related to vascular mortality without any evidence of a threshold. The American Heart Association Stroke Council has also stated that the contribution of BP to ischemic stroke decreases with age (*18*).

Although the reduction of relative risk by lowering BP was smaller in elderly patients than in younger patients in the present study, BP control remains very important for older patients because absolute incidence of CV events is much higher in the older age groups. Ferrucci *et al.* demonstrated the importance of treating isolated systolic hypertension in older patients with a high-risk profile in the Systolic Hypertension in the Elderly Program (SHEP) study, with the number of patients who need to be treated to prevent one CV event becoming progressively smaller for each higher CV risk quartile (*19*). Like the present study, other large-scale clinical trials have demonstrated the benefit of antihypertensive therapy in elderly patients (*20–23*). In addition, clinical guidelines for management of hypertension such as the JNC 7 (*6*) and ESH/ESC (*16*) state that the target BP should be below 140/90 mmHg regardless of age. However, there is little compelling evidence on which to base an optimal target BP for elderly persons (*24*). Detailed analysis of the Hisayama study has demonstrated that the incidence of vascular events increases with rising BP in each risk stratum among younger elderly subjects, but a similar relationship was not observed among the older elderly subjects (*25*). In the SHEP sub-analysis,

reduction of BP to lower than 140 mmHg increased events in elderly hypertensive patients whose BP at entry was higher than 160 mmHg (*26*).

It is still controversial whether BP control for very elderly patients is beneficial as indicated for middle-aged patients. Interestingly, BP was controlled below 140/90 mmHg in about half of our very elderly Japanese hypertensive patients (aged ≥85 years), and they had a low incidence of CV events even after adjustment for other risk factors such as diabetes and a history of cardio-/cerebrovascular disease. However, the baseline SBP was 10 mmHg lower in the BP-controlled group than in the BP-uncontrolled group. This difference of baseline BP may account in part for the difference in the incidence of CV events. The Hypertension in the Very Elderly Trial is ongoing as a prospective randomized open blinded end-points investigation of elderly (>80 years old) hypertensive patients with a target BP <150/80 mmHg (*27*). Preliminary results have demonstrated a reduction of stroke events and stroke mortality (*28*). In Japan, some intervention trials, such as the Japan Trial to Assess Optimal Systolic Blood Pressure in Elderly Hypertensive Patients and the Valsartan in Elderly Isolated Systolic Hypertension study, have been conducted to assess optimal BP for the Japanese elderly patients with hypertension, and these results are expected to provide further information for the management of elderly patients with hypertension (*29, 30*). It will be necessary to investigate and discuss such findings from those interventional studies further before setting an optimal treatment for hypertension in elderly patients.

A J-shaped curve was observed between total mortality and SBP or DBP level in the J-HEALTH study. Death from stroke and MI increased with an elevation of SBP, but a J-shaped relationship was observed between DBP and cardiovascular mortality. Neither SBP nor DBP affected cancer mortality in this study. Boutitie *et al*. reported a J-shaped relationship between risk for death and SBP or DBP in their meta-analysis of seven randomized clinical trials (*31*). They concluded that the increased risk for death observed in patients with low BP was not related to antihypertensive treatment and was not specific to BP-related events. They speculated that the J-shaped curve was probably attributable to poor health conditions that led to low BP and an increased risk for death, and we tend to agree with this assessment. In addition, the mean age was higher in the group of patients with DBP <75 mmHg compared to those with DBP ≥75 mmHg in the present study. Elderly patients were frequently associated with low DBP and poor health conditions, probably resulting in an increased risk for death, although mortality was adjusted for age. We should note that one important limitation of this study was that it was not an intervention trial, so it was difficult to set an optimal BP level.

In summary, we demonstrated that classical risk factors, such as male gender, aging, diabetes mellitus, a history of cardio-/cerebrovascular disease, and smoking, are independent risk factors for future vascular events in daily clinical practice

in Japan. After adjustment for these factors, there was a clear relation between BP control and CV events. The incidence of CV events was significantly increased in patients with BP ≥140/85 mmHg and in diabetic patients with BP ≥130/85 mmHg during treatment, suggesting the validity of adopting current clinical guidelines for Japanese hypertensive patients. Furthermore, about half of our very elderly patients with hypertension had a BP below 140/90 mmHg on losartan-based treatment, and these well-controlled very elderly patients had significantly fewer CV events than uncontrolled patients. In conclusion, there was a clear impact of BP during treatment on CV events in our Japanese hypertensive patients.

Appendix

J-HEALTH Study Committees

Monitoring Committee: Takenori Yamaguchi (Chair), Tanenao Eto, Toshiharu Furukawa, Katsumi Yoshida.

Event Assessment Committee: Hiroaki Naritomi (Chair), Yoichiro Hashimoto, Uichi Ikeda, Mitsuaki Isobe, Toshio Kushiro, Ken Nagata, Kazuyuki Shimada, Takemori Yamawaki.

Safety Assessment Committee: Kendo Kiyosawa (Chair), Hiroshi Hirose, Sadayoshi Ito, Akinori Kasahara, Hiroshi Kawabe, Genjiro Kimura, Hirofumi Makino, Mitsuhiko Moriyama, Ikuo Saito, Hiromichi Suzuki, Eiji Tanaka.

Medical Expert Advisory and Publication Committee: Hiroaki Naritomi (Chair), Toshiro Fujita, Sadayoshi Ito, Toshio Ogihara, Kazuyuki Shimada, Kazuaki Shimamoto, Heizo Tanaka, Nobuo Yoshiike.

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References

1. The ALLHAT Officers and Coordinators for the ALLHAT Collaborative Research Group: Major outcomes in high-risk hypertensive patients randomized to angiotensin-converting enzyme inhibitor or calcium channel blocker *vs* diuretic: the Antihypertensive and Lipid-Lowering Treatment to Prevent Heart Attack Trial (ALLHAT). *JAMA* 2002; **288**: 2981–2997.

2. Dahlof B, Devereux RB, Kjeldsen SE, *et al*: Cardiovascular morbidity and mortality in the Losartan Intervention For Endpoint reduction in hypertension study (LIFE): a randomised trial against atenolol. *Lancet* 2002; **359**: 995–1003.

3. Neal B, MacMahon S, Chapman N, Blood Pressure Lowering Treatment Trialists' Collaboration: Effects of ACE inhibitors, calcium antagonists, and other blood-pressure–lowering drugs: results of prospectively designed overviews of randomised trials. *Lancet* 2000; **356**: 1955–1964.

4. Staessen JA, Wang JG, Thijs L: Cardiovascular protection

and blood pressure reduction: a meta-analysis. *Lancet* 2001; **358**: 1305–1315.

5. Japanese Society of Hypertension: Guidelines for the management of hypertension (JSH 2004). *Hypertens Res* 2006; **29** (Suppl): S1–S105.

6. Chobanian AV, Bakris GL, Black HR, *et al*: Seventh report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. *Hypertension* 2003; **42**: 1206–1252.

7. Naritomi H, Fujita T, Ito S, *et al*: Design and baseline characteristics of a cohort study in Japanese patients with hypertension: Japan Hypertension Evaluation with Angiotensin II Antagonist Losartan Therapy (J-HEALTH). *Hypertens Res* 2007; **30**: 807–814.

8. Naritomi H, Fujita T, Ito S, *et al*: Efficacy and safety of long-term losartan therapy demonstrated by a cohort study in Japanese patients with hypertension: the Japan Hypertension Evaluation with Angiotensin II Antagonist Losartan Therapy (J-HEALTH) Study. *Hypertens Res* 2008; **31**: 295–304.

9. Black HR, Elliott WJ, Grandits G, *et al*: Principal results of the Controlled Onset Verapamil Investigation of Cardiovascular End Points (CONVINCE) trial. *JAMA* 2003; **289**: 2073–2082.

10. Tanizaki Y, Kiyohara Y, Kato I, *et al*: Incidence and risk factors for subtypes of cerebral infarction in a general population: the Hisayama study. *Stroke* 2000; **31**: 2616–2622.

11. Matsuzaki M, Kita T, Mabuchi H, *et al*: Large scale cohort study of the relationship between serum cholesterol concentration and coronary events with low-dose simvastatin therapy in Japanese patients with hypercholesterolemia. *Circ J* 2002; **66**: 1087–1095.

12. Ueda K, Omae T, Hasuo Y, *et al*: Prognosis and outcome of elderly hypertensives in a Japanese community: results from a long-term prospective study. *J Hypertens* 1988; **6**: 991–997.

13. Hansson L, Zanchetti A, Carruthers SG, *et al*, HOT Study Group: Effects of intensive blood-pressure lowering and low-dose aspirin in patients with hypertension: principal results of the Hypertension Optimal Treatment (HOT) randomised trial. *Lancet* 1998; **351**: 1755–1762.

14. Shimamoto K, Kita T, Mabuchi H, *et al*: Effects of hypertension and type 2 diabetes mellitus on the risk of total cardiovascular events in Japanese patients with hypercholesterolemia: Implications from the Japan Lipid Intervention Trial (J-LIT). *Hypertens Res* 2007; **30**: 119–123.

15. American Diabetes Association: Treatment of hypertension in adults with diabetes. *Diabetes Care* 2002; **25**: 199–201.

16. Guidelines Committee: 2003 European Society of Hypertension–European Society of Cardiology guidelines for the management of arterial hypertension. *J Hypertens* 2003; **21**: 1011–1053.

17. Lewington S, Clarke R, Qizilbash N, Pete R, Collins R, Prospective Studies Collaboration: Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. *Lancet* 2002; **360**: 1903–1913.

18. Goldstein LB, Adams R, Becker K, *et al*: Primary prevention of ischemic stroke: a statement for healthcare professionals from the Stroke Council of the American Heart Association. *Circulation* 2001; **103**: 163–182.

19. Ferrucci L, Furberg CD, Penninx BWJH, *et al*: Treatment of isolated systolic hypertension is most effective in older patients with high-risk profile. *Circulation* 2001; **104**: 1923–1926.

20. Amery A, Birkenhäger W, Brixko P, *et al*: Mortality and morbidity results from the European Working Party on High Blood Pressure in the Elderly trial. *Lancet* 1985; **1** (8442): 1349–1354.

21. Dahlof B, Lindholm LH, Hansson L, Schersten B, Ekbom T, Wester PO: Morbidity and mortality in the Swedish Trial in Old Patients with Hypertension (STOP-Hypertension). *Lancet* 1991; **338**: 1281–1285.

22. Heikinheimo RJ, Haavisto MV, Kaarela RH, Kanto AJ, Koivunen MJ, Rajala SA: Blood pressure in the very old. *J Hypertens* 1990; **8**: 361–367.

23. Kjeldsen SE, Dahlof B, Devereux RB, *et al*: Effects of losartan on cardiovascular morbidity and mortality in patients with isolated systolic hypertension and left ventricular hypertrophy: a Losartan Intervention For Endpoint Reduction (LIFE) substudy. *JAMA* 2002; **288**: 1491–1498.

24. August P: Initial treatment of hypertension. *N Engl J Med* 2003; **348**: 610–617.

25. Arima H, Tanizaki Y, Kiyohara Y, *et al*: Validity of the JNC VI recommendations for the management of hypertension in a general population of Japanese elderly: the Hisayama study. *Arch Intern Med* 2003; **163**: 361–366.

26. Perry HM Jr, Davis BR, Price TR, *et al*: Effect of treating isolated systolic hypertension on the risk of developing various types and subtypes of stroke: the Systolic Hypertension in the Elderly Program (SHEP). *JAMA* 2000; **284**: 465–471.

27. Bulpitt C, Fletcher A, Beckett N, *et al*: Hypertension in the Very Elderly Trial (HYVET): protocol for the main trial. *Drugs Aging* 2001; **18**: 151–164.

28. Bulpitt CJ, Beckett NS, Cooke J, *et al*: Results of the pilot study for the Hypertension in the Very Elderly Trial. *J Hypertens* 2003; **21**: 2409–2417.

29. JATOS Study Group: The Japanese Trial to Assess Optimal Systolic Blood Pressure in Elderly Hypertensive Patients (JATOS): protocol, patient characteristics, and blood pressure during the first 12 months. *Hypertens Res* 2005; **28**: 513–520.

30. Ogihara T, Saruta T, Matsuoka H, *et al*: Valsartan in Elderly Isolated Systolic Hypertension (VALISH) study: rationale and design. *Hypertens Res* 2004; **27**: 657–661.

31. Boutitie F, Gueyffier F, Pocock S, Fagard R, Boissel JP, The INDANA Project Steering Committee: J-shaped relationship between blood pressure and mortality in hypertensive patients: new insights from a meta-analysis of individual-patient data. *Ann Intern Med* 2002; **136**: 438–448.

Original Paper

Quantitative Evaluation of Carotid Plaque Echogenicity by Integrated Backscatter Analysis: Correlation with Symptomatic History and Histologic Findings

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Key Words

Abstract

Background and Purpose: Echogenicity of carotid plaque well reflects the risk of ischemic stroke and may be predictive of the histologic content of the plaque. However, objective evaluation of plaque echogenicity has been hampered by a lack of established quantitative measures. This study examined the relation between echogenicity assessed by integrated backscatter (IBS) analysis and (1) symptomatic history and (2) histologic features of carotid plaques. **Methods:** We used acoustic densitometry to quantify by IBS analysis the echogenicity of 31 carotid plaques of 26 patients under-going carotid endarterectomy or stenting. IBS was subsequently compared with histologic findings of the respective tissue in 10 patients who underwent endarterectomy. The IBS value was calibrated with 2 reference structures (vessel lumen and adventitia) as the IBS index. **Results:** The IBS index of symptomatic plaques was lower than that of asymptomatic plaques (23.1 ± 12.5 vs. 36.5 ± 18.2, $p < 0.05$). The IBS index in fatty/necrotic atheromatous sites ($n = 20$, 16.6 ± 10.7) was lower than that in fibrous ($n = 26$, 42.4 ± 13.6, $p < 0.01$) or calcified ($n = 11$, 87.7 ± 17.4, $p < 0.01$) sites and the same as that in intraplaque hemorrhagic sites ($n = 50$, 23.6 ± 16.9). **Conclusions:** Carotid plaque echogenicity, as quantitatively assessed by IBS analysis, correlates well with the presence or absence of prior symptoms and histologic contents of the plaques. IBS analysis may aid in the assessment of carotid plaque-related risk of stroke.

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Introduction

Carotid atherosclerosis with echolucent plaque is closely related to the occurrence of cerebral infarction [1, 2]. Therefore, determining not only the severity of carotid stenosis but also the features of the plaque by means of high-resolution ultrasound is important for the stratification of high-risk patients with incident stroke [3, 4]. Several classifications of plaque morphology used thus far are subjective and qualitative [5–10]. Recently, two standardized and quantitative methods for the assessment of plaque echogenicity have been introduced: gray-scale median [11–13] and integrated backscatter (IBS) analyses [14–16]. The measurement of IBS is based on the analysis of unprocessed radiofrequency signals to derive quantitative ultrasonic indexes. Several studies have examined IBS and histologic features of carotid plaque [14–18]. Takiuchi et al. [14] reported a low IBS value in atheromatous lesions in freshly excised human aorta. Both Urbani et al. [15] and Waki et al. [16] also reported low IBS values in lipid and hemorrhagic lesions in carotid specimens obtained from symptomatic patients who underwent carotid endarterectomy. On the basis of these findings, several laboratories have investigated the relation of risk factors to carotid plaque echogenicity with the use of IBS [19, 20]. We defined the IBS index of the carotid plaque by normalizing it with the 2 reference points (blood and adventitia) and found an association between the serum interleukin-6 level and the IBS index [21]. Although clinical and histologic characteristics have been studied extensively in terms of carotid plaque echogenicity with gray-scale median analysis [22–24], few reported studies have examined the clinical and histologic value of IBS analysis. To strengthen the clinical value of the IBS index, we attempted to clarify the association of the IBS index with clinical symptoms in patients with severe carotid stenosis and with histologic findings in both symptomatic and asymptomatic patients who underwent carotid endarterectomy.

Subjects and Methods

Subjects

Between March 2000 and January 2001, 31 carotid plaques from 26 patients, all of whom were admitted to our institution for carotid endarterectomy or carotid stenting, were studied. Twelve plaques caused no ipsilateral symptoms, and 19 caused symptoms including ischemic stroke, transient ischemic attack, and amaurosis fugax. Five patients had ipsilateral symptomatic plaques and contralateral asymptomatic plaques. Preoperative angiography was routinely performed in all cases to determine the surgical indication. The degree of internal carotid artery stenosis was assessed by conventional arteriography according to NASCET criteria [25]. The indication for carotid endarterectomy or stenting was internal carotid artery stenosis of >70% or 50–69% with repeated ischemic cerebrovascular events or severe ulcerative atheroma. Of the 26 patients, 22 were men and 4 were women. Mean age ± SD was 69.1 ± 6.2 years (range, 55–78 years). Information obtained for each patient included age, sex, referring diagnosis, smoking status, current medical therapy, and history of hypertension, diabetes mellitus, hyperlipidemia, or ischemic heart disease. The overall average degree of internal carotid artery stenosis determined angiographically was 83.5 ± 14.5% (range, 50–99%). Seventeen patients underwent carotid endarterectomy and 14 patients underwent carotid stenting. We obtained informed consent from all patients.

Carotid Ultrasonography

Conventional carotid duplex ultrasonography was performed before carotid endarterectomy or stenting. All ultrasound scans and subsequent analyses were performed with a Phillips SONOS 5500 system equipped with a 7.5-MHz linear-array transducer. Initially, the common and internal carotid arteries were scanned cross-sectionally and longitudinally by B-mode methods through which the largest plaque was identified for evaluation of plaque echogenicity. The corresponding colored image was also digitized, an essential requirement for outlining echolucent plaques, with their hypoechoic component adjacent to the lumen of the vessel. Subsequently, with the use of acoustic densitometry, 20 longitudinal IBS images per the largest plaque were acquired into cine-loop memory and stored on optical disks. The acoustic densitometry system is capable of providing 2-dimensional IBS images in which the gray level is displayed proportionally to the integrated backscattered power. The IBS values were obtained from the plaque (pl), vessel lumen (lm), and adventitia (ad) at the same depth of the plaque. Because reproducibility of carotid plaque echogenicity was better when 2 reference structures (lm and ad) were used compared to 1, we defined the IBS index as: $(pl - lm)/(ad - lm) \times 100$. Accordingly, a low IBS index corresponds to low echogenicity. For all plaques in the study, the region of interest was placed in the whole plaque (fig. 1). For comparison with histologic findings in 10 plaque specimens for which a good-quality ultrasonic study was completed, transverse IBS images were acquired at intervals of 5 mm from the carotid bifurcation to the internal carotid artery, corresponding to the sites in the histologic specimens (fig. 2, 3). A region of interest of 11×11 pixels was placed in the plaque. A total of 107 discrete regions in 10 plaques were examined and analyzed.

We have previously assessed intraobserver and interobserver reproducibility of variation for IBS index measurements with acceptable results (8.9 and 9.2%, respectively).

MRI Study

Diffusion-weighted MRI (DWI) was performed before the surgical or endovascular procedure to detect recent ischemic lesions in all patients. The overall mean interval between the MRI and ultrasonography was 8.0 ± 7.9 days (range, 1–35 days). Twenty-three of all patients underwent MRI within 14 days before ultrasonography. Twenty-eight carotid plaques from 23 patients were evaluated by DWI to involve new infarcts that were related to the ipsilateral carotid stenosis.

Fig. 1. IBS analysis of carotid plaques. Using acoustic densitometry, IBS values are obtained from the whole plaque (pl), vessel lumen (lm), and adventitia (ad) at the same depth, where the IBS index is defined as: (pl − lm)/(ad − lm) × 100.
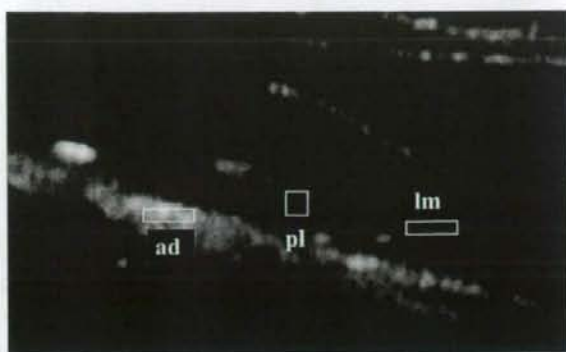
Fig. 2. IBS analysis of carotid plaques for comparison with histologic findings. Note the location of the region of interest (1.1 × 1.1 pixel) in the area of the plaque. Using acoustic densitometry, IBS values are obtained from the plaque (pl), vessel lumen (lm), and adventitia (ad), where the IBS index is defined as: (pl − lm)/(ad − lm) × 100.
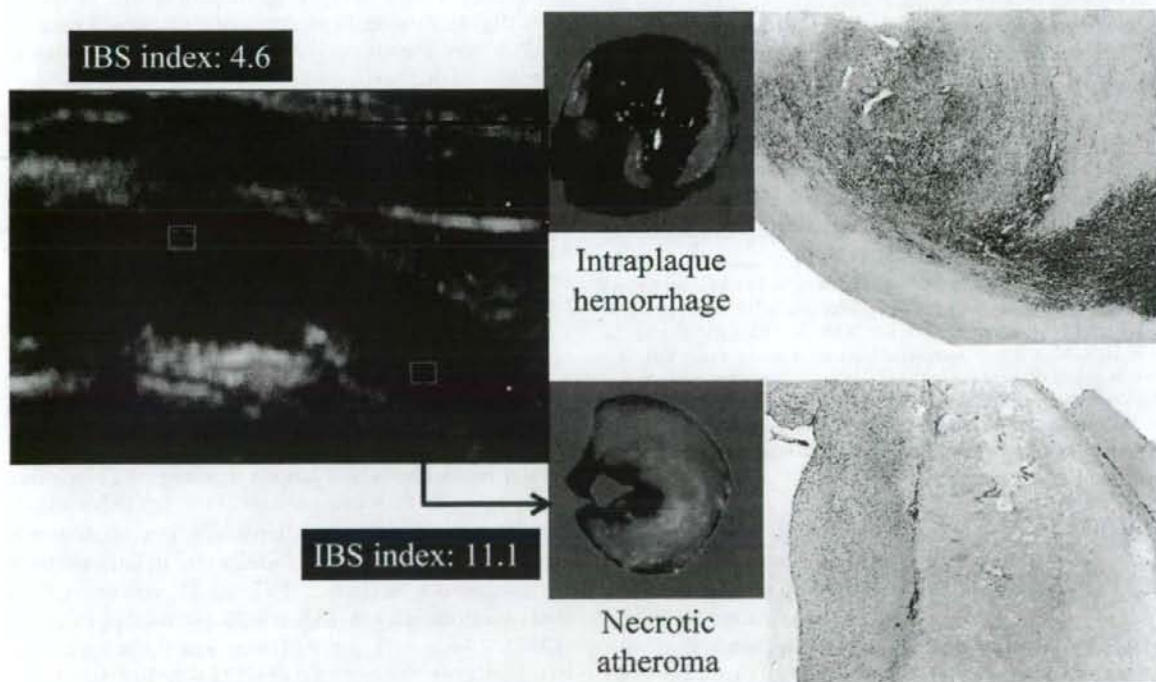
Fig. 3. Histologic findings in the carotid specimens. Left = IBS image of carotid stenosis and IBS indexes corresponding to the plaque specimens; middle = endarterectomy specimen pathology; right = high-power photomicrographs of the plaque specimen with recent intraplaque hemorrhage (top) and necrotic atheroma (bottom; hematoxylin and eosin stain; original magnification ×15). IBS index is 4.6 in the intraplaque hemorrhage site and 11.1 in the necrotic atheroma site.

Table 1. Baseline characteristics of subjects

	Symptomatic	Asymptomatic	p value
Number	19	7	–
Age (mean ± SD), years	68.5±6.9	69.6±5.1	0.79
Male, %	79	100	0.55
Hypertension, %	68	71	0.99
Diabetes mellitus, %	21	14	0.99
Hyperlipidemia, %	53	71	0.66
History of CVD, %	31	29	0.99
Smokers, %	79	86	0.99
Statin use, %	26	29	0.99
Aspirin use, %	47	71	0.39
Stenosis severity, %	87.1±13.7	80.3±12.0	0.10

Age and stenosis severity are compared using the Mann-Whitney U test. The percentage of males, hypertension, diabetes mellitus, hyperlipidemia, history of CVD, smokers, statin use and aspirin use are compared using Fisher's exact test between the symptomatic and asymptomatic plaques. CVD = Cardiovascular disease.

Fig. 4. The IBS index of symptomatic and asymptomatic carotid plaques. The IBS index of symptomatic plaques is lower than that of asymptomatic plaques (23.1 ± 12.5 vs. 36.5 ± 18.2).

Histological Analysis

Samples of the 10 plaque specimens were fixed in 10% formalin. They were embedded in paraffin, and 5-μm sections of each sample were studied histologically with hematoxylin and eosin and Masson's trichrome stains. The microscopic sections were analyzed by an experienced pathologist blinded to the echographic results. Each plaque sample was evaluated and classified into 1 of 4 categories: (1) intraplaque hemorrhage (fig. 3), i.e., leakage of erythrocytes inside the plaque without fibrin mesh; (2) fibrous tissue, i.e., presence of fibrous tissue, collagen bundles, and smooth muscle cells; (3) atheromatous tissue specimen (fig. 3), i.e., a necrotic core or accumulation of lipid, and (4) calcified tissue.
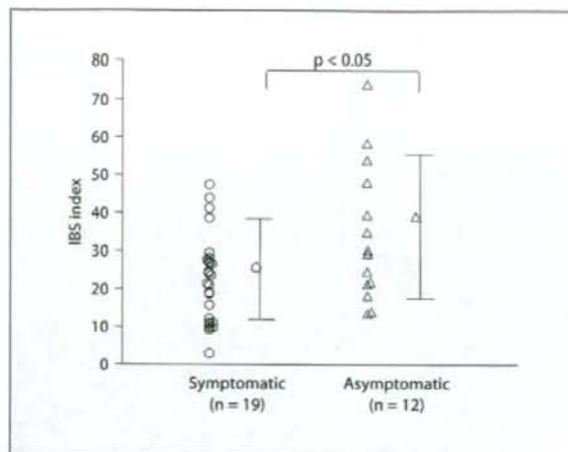
Statistical Analysis

Values are shown as mean ± SD. Differences between the symptomatic and asymptomatic groups were assessed using the Mann-Whitney U test for continuous variables and Fisher's exact test for categorical variables. Differences in the frequency of DWI-positive lesions between the ≤30 IBS index group and the >30 IBS index group were analyzed by Fisher's exact test. Between-group differences in the IBS index values were subjected to ANOVA with Scheffé's multiple comparison test.

All analyses were performed with StatView software. A probability value of <0.05 was considered significant.

**Results**

Clinical characteristics of the study patients are shown in table 1. There were no significant differences in age, sex, risk factors for stroke, history of ischemic heart disease, therapeutic variables, and degree of carotid stenosis between patients. The IBS index of symptomatic plaques was lower than that of asymptomatic plaques (23.1 ± 12.5 vs. 36.5 ± 18.2, p < 0.05), whereas the degree of stenosis was the same irrespective of the presence of symptoms (fig. 4). Among 16 symptomatic plaques, 5 plaques (31.3%) were shown by DWI to involve recent infarcts that were related to the ipsilateral carotid stenosis, whereas 2 plaques (16.7%) among 12 asymptomatic plaques were shown to involve recent infarcts. All infarcts shown by DWI were localized in the carotid artery territory and the type of infarcts was atheroembolism. Among the 5 symptomatic plaques with new lesions, 4 plaques showed a low IBS index (≤30) and 1 plaque a high IBS index (>30). Among the 2 asymptomatic plaques with new lesions, all of them showed a low IBS index of 10.6 and 20.7. In the subgroup in which the IBS index was ≤30, 6 of 16 (37.5%) plaques were shown by DWI to involve new infarcts, whereas in the subgroup in which the IBS index was >30, only 1 of 12 (8%) plaques was shown to involve new infarcts. The ≤30 IBS index group tended to show a greater incidence of new lesions than the >30 IBS index group (p = 0.077). A comparison of the IBS index values for plaques with different microscopic characteristics is shown in figure 5. The IBS index value in fatty/necrotic atheromatous sites (16.6 ± 10.7, n = 20) was lower than that in fibrous (42.4 ± 13.6, n = 26, p < 0.01) or calcified (87.7 ± 17.4, n = 11, p < 0.01) sites and the same as that in intraplaque hemorrhagic sites (23.6 ± 16.9, n = 50).

Quantitative Evaluation of Carotid
Plaque Echogenicity by IBS Analysis
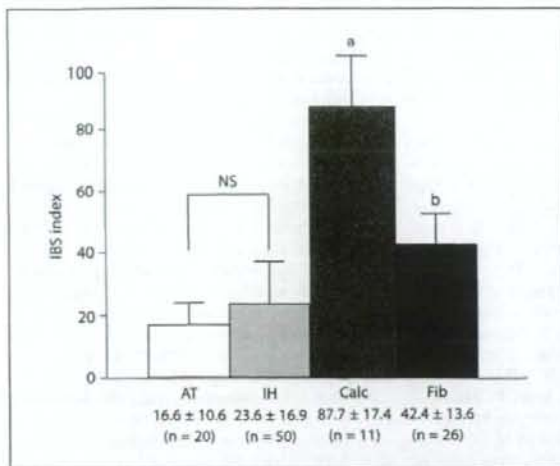
Fig. 5. Comparison of the IBS index with histologic features of carotid plaques. The IBS index in fatty/necrotic atheromatous sites was lower than that in fibrous or calcified sites and the same as that in intraplaque hemorrhagic sites. AT = Atheroma; IH = intraplaque hemorrhage; Calc = calcified; Fib = fibrous. [a] p < 0.05 vs. all other groups, [b] p < 0.05 vs. AT and IH.

Discussion

In this study, we showed that a low IBS index was associated with a history of ischemic stroke and lipid and hemorrhagic lesions within the plaque. These findings are in good agreement with previously reported findings based on IBS [14–18] and gray-scale median analyses [11–13, 22–24], suggesting the clinical value of the IBS index for determining plaque vulnerability. Furthermore, using DWI, we observed that carotid plaque with a low IBS index tended to be associated with new ischemic lesions. The present findings support the use of the IBS index to discover factors related to plaque vulnerability. In our follow-up study for carotid atherosclerosis [26], Yamagami et al. [21] confirmed the association between a low IBS index and risk factors such as smoking and dyslipidemia and showed that increased blood IL-6 levels are related to a low IBS index, pointing to the role of inflammation in plaque vulnerability. The IBS index analysis may allow examination of the involvement of genetic factors and gene polymorphism in plaque instability; such involvement has rarely been examined but is undoubtedly important. Furthermore, the present results may support the use of the IBS index as a surrogate marker of plaque instability in longitudinal interventional studies. Wata-

nabe et al. [20] report on the effect of 1-year treatmen with pravastatin on plaque echogenicity, suggesting stabilization of carotid atheroma by statin treatment. The anti-atherogenic effect of other statins, renin-angiotensin system inhibitors and insulin-sensitizing drugs coulc be examined clinically by the use of the IBS index. Interpretation of our findings is limited. First, it remains unclear how the IBS index is best assessed for heterogeneou: mixed plaque. Waki et al. [16] showed that the IBS value measured at the interface of the plaque reflects the thickness of the fibrous cap. Further studies are needed to clarify the significance of regional IBS indexes within a plaque. Second, the IBS index did not differ between atheroma and intraplaque hemorrhage. All previous studie: with carotid ultrasonography, whether qualitative, grayscale median, or IBS analyses, showed the same result [7 8, 14, 15, 27]. So far, we have been unable to overcome thi: problem. Fortunately, both atheroma and intraplaque hemorrhage are signs of unstable plaque; therefore, thi: problem may not hamper the use of IBS for evaluating plaque instability. Third, the IBS index of symptomatic plaques was lower than that of asymptomatic plaques, bu there is an overlap of the individual IBS index in symptomatic and asymptomatic plaques. This means that it i: difficult to discriminate between stable symptomatic plaques which have healed or evolved since the time o neurologic symptoms, and unstable asymptomatic plaques with asymptomatic new lesions in DWI. Fourth IBS can be measured only with certain ultrasonic device: [14, 16–20]. Comparison of the clinical and histologic significance of IBS and gray-scale median analysis will clarify whether IBS is better and more valuable than grayscale median analysis to assess plaque echogenicity.

In conclusion, we showed that the IBS index is closely associated with clinical symptoms and histologic feature: in patients with severe carotid stenosis. Future longitudinal studies are needed to clarify the association of the IBS index in carotid plaque with incident cardiovasculai events and plaque progression.

References

1 Polak JF, Shemanski L, O'Leary DH, Lefkowitz D, Prince TR, Savage PJ, Brant WE, Reic C: Hypoechoic plaque at US of the carotic artery: an independent risk factor for incident stroke in adults aged 65 years or older Radiology 1998;208:649–654.
2 Mathiesen EB, Bønaa KH, Joakimsen O Echolucent plaques are associated with high risk of ischemic cerebrovascular events in carotid stenosis: the tromsø study. Circulation 2001;103:2171–2175.

Grønholdt ML, Nordestgaard BG, Schroeder TV, Vorstrup S, Sillesen H: Ultrasonic echolucent carotid plaques predict future strokes. Circulation 2001;104:68–73.

Hennerici M, Baezner H, Daffertshofer M: Ultrasound and arterial disease. Cerebrovasc Dis 2004;17(suppl 1):19–33.

Fisher M, Paganini-Hill A, Martin A, Cosgrove M, Toole JF, Barnett HJ, Norris J: Carotid plaque pathology: thrombosis, ulceration, and stroke pathogenesis. Stroke 2005; 36:253–257.

Bassiouny HS, Sakaguchi Y, Mikucki SA, McKinsey JF, Piano G, Gewertz BL, Glagov S: Juxtalumenal location of plaque necrosis and neoformation in symptomatic carotid stenosis. J Vasc Surg 1997;26:585–594.

Gray-Weale AC, Graham JC, Burnett JR, Byrne K, Lusby RJ: Carotid artery atheroma: comparison of preoperative B-mode ultrasound appearance with carotid endarterectomy specimen pathology. J Cardiovasc Surg (Torino) 1988;29:676–681.

Hatsukami TS, Ferguson MS, Beach KW, Gordon D, Detmer P, Burns D: Carotid plaque morphology and clinical events. Stroke 1997;28:95–100.

Carr S, Farb A, Pearce WH, Virmani R, Yao JS: Atherosclerotic plaque rupture in symptomatic carotid artery stenosis. J Vasc Surg 1996;23:755–765.

Uwatoko T, Toyoda K, Inoue T, Yasumori K, Hirai Y, Makihara N, Fujimoto S, Ibayashi S, Iida M, Okada Y: Carotid artery calcification on multislice detector-row computed tomography. Cerebrovasc Dis 2007;24:20–26.

el-Barghouty N, Nicolaides A, Bahal V, Geroulakos G, Androulakis A: The identification of the high risk carotid plaque. Eur J Vasc Endovasc Surg 1996;11:470–478.

Sabetai MM, Tegos TJ, Nicolaides AN, Dhanjil S, Pare GJ, Stevens JM: Reproducibility of computer-quantified carotid plaque echogenicity. Can we overcome the subjectivity? Stroke 2000;31:2189–2196.

13 Lal BK, Hobson RW 2nd, Pappas PJ, Kubicka R, Hameed M, Chakhtoura EY, Jamil Z, Padberg FT Jr, Haser PB, Durán WN: Pixel distribution analysis of B-mode ultrasound scan images predicts histologic features of atherosclerotic carotid plaques. J Vasc Surg 2002;35:1210–1217.

14 Takiuchi S, Rakugi H, Honda K, Masuyama T, Hirata N, Ito H, Sugimoto K, Yanagitani Y, Moriguchi K, Okamura A, Higaki J, Ogihara T: Quantitative ultrasonic tissue characterization can identify high-risk atherosclerotic alteration in human carotid arteries. Circulation 2000;102:766–770.

15 Urbani MP, Picano E, Parenti G, Mazzarisi A, Fiori L, Paterni M, Pelosi G, Landini L: In vivo radiofrequency-based ultrasonic tissue characterization of the atherosclerotic plaque. Stroke 1993;24:1507–1512.

16 Waki H, Masuyama T, Mori H, Maeda T, Kitade K, Moriyasu K, Tsujimoto M, Fujimoto K, Koshimae N, Matsuura N: Ultrasonic tissue characterization of the atherosclerotic carotid artery histological correlates of carotid integrated backscatter. Circ J 2003;67: 1013–1016.

17 Waters KR, Bridal SL, Cohen-Bacrie C, Levrier C, Fornès P, Laugier P: Parametric analysis of carotid plaque using a clinical ultrasound imaging system. Ultrasound Med Biol 2003;29:1521–1530.

18 Kawasaki M, Takatsu H, Noda T, Ito Y, Kunishima A, Arai M, Nishigaki K, Takemura G, Morita N, Minatoguchi S, Fujiwara H: Noninvasive quantitative tissue characterization and two-dimensional color-coded map of human atherosclerotic lesions using ultrasound integrated backscatter: comparison between histology and integrated backscatter images. J Am Coll Cardiol 2001;38:486–492.

19 Katakami N, Kaneto H, Matsuhisa M, Umayahara Y, Kosugi K, Yamasaki Y: Clustering of several cardiovascular risk factors affects tissue characteristics of the carotid artery. Atherosclerosis 2008;198:208–213.

20 Watanabe K, Sugiyama S, Kugiyama K, Honda O, Fukushima H, Koga H, Horibata Y, Hirai T, Sakamoto T, Yoshimura M, Yamashita Y, Ogawa H: Stabilization of carotid atheroma assessed by quantitative ultrasound analysis in nonhypercholesterolemic patients with coronary artery disease. J Am Coll Cardiol 2005;46:2022–2030.

21 Yamagami H, Kitagawa K, Nagai Y, Hougaku H, Sakaguchi M, Kuwabara K, Kondo K, Masuyama T, Matsumot M, Hori M: Higher levels of interleukin-6 are associated with lower echogenicity of carotid artery plaques. Stroke 2004;35:677–681.

22 Tegos TJ, Sohail M, Sabetai MM, Robless P, Akbar N, Pare G, Stansby G, Nicolaides AN: Echomorphologic and histopathologic characteristics of unstable carotid plaques. Am J Neuroradiol 2000;21:1937–1944.

23 Grønholdt MLM, Nordestgaard BG, Wiebe BM, Wilhjelm JE, Sillesen H: Echo-lucency of computerized ultrasound images of carotid atherosclerotic plaques are associated with increased levels of triglyceride-rich lipoproteins as well as increased plaque lipid content. Circulation 1998;97:34–40.

24 Sabetai MM, Tegos TJ, Nicolaides AN, El-Atrozy TS, Dhanjil S, Griffin M, Belcaro G, Geroulakos G: Hemispheric symptoms and carotid plaque echomorphology. J Vasc Surg 2000;31:39S–49S.

25 North American Symptomatic Carotid Endarterectomy Trial Collaborators: Beneficial effect of carotid endarterectomy in symptomatic patients with high-grade carotid stenosis. N Engl Med 1991;325:445–453.

26 Kitagawa K, Hougaku H, Yamagami H, Hashimoto H, Itoh T, Shimizu Y, Takahashi D, Murata S, Seike Y, Kondo K, Hoshi T, Furukado S, Abe Y, Yagita Y, Sakaguchi M, Tagaya M, Etani H, Fukunaga R, Nagani Y, Matsumoto M, Hori M: Carotid intima-media thickness and risk of cardiovascular events in high-risk patients. Cerebrovasc Dis 2007;24:35–42.

27 Sztajzel R, Momjian S, Momjian-Mayor I, Murith N, Djebaili K, Boissard G, Comelli M, Pizolatto G: Stratified gray-scale median analysis and color mapping of the carotid plaque: correlation with endarterectomy specimen histology of 28 patients. Stroke 2005;36:741–745.

Heparin-Induced Thrombocytopenia: A Serious Complication of Heparin Therapy for Acute Stroke

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Abstract
Background: Despite the lack of supporting evidence, un-
fractionated heparin (UFH) is frequently given to acute isch-
emic stroke patients. This study was designed to determine
the incidence of heparin-induced thrombocytopenia (HIT)
during acute stroke and to elucidate the clinical features of
stroke patients with HIT. **Methods:** Of 1,078 consecutive pa-
tients with acute ischemic stroke, 392 were given intrave-
nous UFH. Ten of these developed prominent thrombocyto-
penia without any other underlying etiology; they were
suspected of having HIT. These 10 patients were studied ret-
rospectively. The clinical diagnosis of HIT was made accord-
ing to two published scoring systems. Antiplatelet factor 4/
heparin antibodies in the plasma were detected by the en-
zyme-linked immunosorbent assay (ELISA) and were con-
firmed by the ^{14}C-serotonin release assay. **Results:** Eight
patients met the criteria for clinical HIT according to both
scoring systems. Of these, serological tests were positive in
2 patients only on ELISA and in 2 patients on both assays. The
amount of UFH given was greater in the 4 patients with pos-
itive serological findings than in the others (p = 0.043). Three
patients developed further thromboembolic events, includ-
ing 1 patient who developed possible cancer-associated
thrombosis. Two patients were dead and the remaining 6
patients were dependent at the time of hospital discharge.
The clinical severity and outcome of these patients were rel-
atively unfavorable compared to other acute patients. **Con-
clusions:** The prevalence of HIT was 0.5% based on both the
clinical scoring systems and serological assays. Monitoring
for HIT should be included in the medical management of
stroke to avoid further complications.

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Introduction

Heparin-induced thrombocytopenia (HIT) is caused
by platelet-activating IgG antibodies that recognize plate-
let factor 4 bound to heparin (anti-PF4/heparin Abs) [1].
It is characterized by thrombocytopenia with an in-
creased risk of thrombosis. Prospective trials have indi-
cated that between 0.3 and 5% of patients treated with
unfractionated heparin (UFH) developed HIT, depend-

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ing on the clinical setting [2–5]. Thrombotic complications (HIT thrombosis syndrome) are frequently associated with HIT, occurring in approximately one third to one half of HIT patients [1].

Some guidelines do not recommend prescribing heparin in acute stroke, and some recommend it mainly for reducing deep vein thrombosis (DVT) and pulmonary embolism (PE) [6–8]. At the National Cardiovascular Center (Osaka, Japan), in addition to being administered to prevent DVT and PE during the acute phase of stroke, UFH is given to: acute stroke patients having embologenic heart diseases or superimposed thrombi on the carotid plaque to prevent embolic complications; patients with particular stroke etiologies, including cerebral arterial dissection and vasculitis, and patients with embolic stroke of unknown origin until the presence of heart diseases is excluded using prolonged electrocardiography and transesophageal echocardiography [9]. A previous study of 137 stroke patients treated with UFH reported that 21 patients (15.3%) developed thrombocytopenia, and 5 of these 21 patients had a recurrent ischemic stroke [10]. In another study, of 18 patients who developed carotid arterial occlusion following endarterectomy, 6 (33%) had an associated heparin-induced coagulation disorder [11]. A recent study of 200 neurological patients treated with UFH for at least 5 days, including 102 patients with cerebrovascular disorders, demonstrated that 41 patients (20.5%) had anti-PF4/heparin Abs and 5 (2.5%) developed HIT, when an enzyme-linked immunosorbent assay (ELISA) was used [12]. However, in other studies based on more stroke patients, no one developed thrombocytopenia during treatment with UFH [13, 14]. The incidence of HIT during acute stroke in the Asian population has not been determined.

In our center, HIT has been recognized as a serious complication of cerebrovascular and cardiovascular diseases for the last several years [15]. Patients who develop thrombocytopenia or thromboembolic events without another underlying etiology during or after UFH therapy are assessed for anti-PF4/heparin Abs using ELISA; if these patients are strongly suspected of having HIT, they are primarily treated with argatroban, a direct thrombin inhibitor. The goal of the present study was to evaluate the incidence and clinical features of HIT patients in a stroke center during and after UFH treatment for acute ischemic stroke. The diagnosis of HIT was made using two clinical scoring systems and two serological tests [ELISA and the ^{14}C-serotonin release assay (SRA)]. In particular, SRA has been recognized to have high sensitivity and specificity for the diagnostic of HIT.

Methods

Between July 2003 and May 2006, 1,078 patients were admitted to the stroke care unit of the National Cardiovascular Center within 7 days after the onset of acute ischemic stroke; 392 of these patients were given UFH during the acute stage. UFH was principally given as a continuous intravenous injection at a rate to maintain an activated partial thromboplastin time 1.5–2.0 times the control values. The data of patients who developed prominent thrombocytopenia (defined as a minimum platelet count <100,000/μl or <60% of the baseline count) within 40 days after the initiation of UFH therapy without any other underlying etiology (such as drug reactions other than heparin and severe infections) were retrospectively analyzed.

The clinical diagnosis of HIT was made according to two scoring systems. The first system, the '4Ts' [16], is composed of four clinical features that are given scores of 0, 1, or 2 for each feature: acute thrombocytopenia; thrombosis or other sequelae; timing of platelet count fall or sequelae, and other cause of thrombocytopenia not evident. Based on the test score, the estimated pretest probabilities of HIT are: low risk (0–3), intermediate risk (4–5), or high risk (6–8). The second system proposed by Pouplard et al. [17] assesses the course of the platelet count (–2, –1, 0, or 2), thrombotic complications including brain infarction (1, 2, or 4), thrombocytopenia after rechallenge with heparin (if done, –1, 0, or 6), and other causes of thrombocytopenia (–2, 0, or 2); based on their total score, patients are categorized into 4 clinical groups: definite (≥6), probable (3–5), possible (1–2), and unlikely HIT (≤0). In the present study, patients who had a high or intermediate score in the first system (4Ts) and a definite, probable, or possible score in the second system were diagnosed as having clinical HIT. In a previous study, patients with a low score according to 4Ts were unlikely to test positive for platelet-activating HIT antibodies including SRA [18]. In another one, patients with 'unlikely HIT' according to the system by Pouplard et al. [19] were unlikely to test positive for platelet aggregation test or SRA. Thus, for patients having clinical HIT, those who met either low score in 4Ts or unlikely HIT in the second system were thought to be ineligible.

In addition, all patients had serological tests for anti-PF4/heparin Abs detected by ELISA (Asserachrom HPIA, Diagnostica Stago, Asnières, France) just after their attending physician suspected that they had HIT. ELISA was done according to the manufacturer's instructions. The titers of the samples were expressed as values of optical density. The result was considered as positive when the titer was greater than the cutoff value, which was determined using the reference control given for each kit. SRA was done to confirm the diagnosis of HIT retrospectively and was performed as described elsewhere at the Loyola University Medical Center (Maywood, Ill., USA) [20]. For each sample, SRA was done using washed platelets from at least 2 donors, known to be reactive in the assay. Positive samples gave ≥20% release of ^{14}C-serotonin in response to 0.1 U/ml heparin (final concentration), with inhibition of that response in the presence of 100 U/ml heparin (final concentration). ELISA and SRA were done by different examiners who were blinded to the clinical information and results of each other assay.

Based on the laboratory results, patients were categorized into three groups as follows: (1) definite HIT (positive results in both ELISA and SRA); (2) suspected HIT (positive result in ELISA, but negative in SRA), and (3) clinically suspected HIT (clinical HIT

Table 1. Diagnosis of clinical and serological HIT

Pa-tient No.	Final diagnosis	First system (4Ts)						Second system (by Pouplard et al.)						ELISA (optical density)[1]	SRA
		1°	2°	3°	4°	score	assessment	1°	2°	3°	4°	score	assessment		
1	definite HIT	1	2	0	2	5	intermediate	2	0	–	2	4	probable	pos. (0.675)	pos.
2	definite HIT	1	2	0	2	5	intermediate	–	–	–	2	2	possible	pos. (0.777)	pos.
3	suspected HIT	2	1	1	1	5	intermediate	2	2	6	–2	8	definite	pos. (0.760)	neg.
4	suspected HIT	2	1	0	1	4	intermediate	2	0	6	0	8	definite	pos. (0.918)	neg.
5	clinically suspected HIT	2	2	1	2	7	high	0	2	0	2	4	probable	neg. (0.177)	neg.
6	clinically suspected HIT	2	0	0	2	4	intermediate	2	–	–	2	4	probable	neg. (0.173)	neg.
7	clinically suspected HIT	1	2	0	2	5	intermediate	3	0	–	1	4	probable	neg. (0.132)	neg.
8	clinically suspected HIT	2	0	1	2	5	intermediate	–1	2	–1	2	2	possible	neg. (0.141)	neg.
9	not HIT	2	2	0	1	5	intermediate	–2	2	–	–2	–2	unlikely	neg. (0.085)	neg.
10	not HIT	2	0	0	1	3	low	–2	0	0	–2	–4	unlikely	neg. (0.111)	neg.

First system (4Ts): 1° = Acute thrombocytopenia; 2° = timing of platelet count fall, thrombosis, or other sequelae; 3° = thrombosis or other sequelae; 4° = other cause of thrombocytopenia not evident. Second system (by Pouplard et al.): 1° = Course of platelet count; 2° = thrombotic complication; 3° = thrombocytopenia after rechallenge of heparin; 4° = other causes of thrombocytopenia. pos. = Positive; neg. = negative.
[1] Cutoff value ranges between 0.473 and 0.560.

according to the scoring systems with negative results in both ELISA and SRA).

The following patient characteristics were obtained from the patients' medical records: age, gender, platelet counts, vascular risk factors such as hypertension, diabetes mellitus, hyperlipidemia, current and past smoking habit, and drinking habit (including occasional drinking). Other risk factors for stroke, such as embologenic heart diseases including atrial fibrillation, were assessed based on the criteria of the Trial of Org 10172 in Acute Stroke Treatment (TOAST) study [21]. Based on the neurological, radiological, cardiological, and hematological profiles, the stroke subtype was determined according to the TOAST subtype classification system by a consensus of stroke neurologists [21]. The neurological severity of each patient was assessed by an experienced stroke neurologist according to the National Institutes of Health Stroke Scale (NIHSS) score on admission; at discharge, activity of daily living was assessed using the modified Rankin scale (mRS).

The duration and dose of UFH, the change in platelet count, and alternative anticoagulant therapy for HIT (if done) were also assessed. The NIHSS and mRS scores were compared between stroke patients treated with UFH who had and those who did not have HIT. The variables between the groups were compared using the Mann-Whitney U test.

Results

During the study period, 10 stroke patients were suspected of having developed HIT since they had thrombocytopenia without any underlying etiology other than heparin exposure. Of these patients, 1 had a high score and 8 had intermediate scores on the first scoring system (4Ts); using the second scoring system, 2 met the criteria of definite, 4 of probable, and 2 of possible HIT (table 1). In particular, 2 patients had a high score using the second scoring system because they developed repeated thrombocytopenia after UFH rechallenge during the chronic stage of stroke. One patient (No. 3) was given small amounts of UFH flushes after the initial thrombocytopenic event to maintain intravenous catheter patency; anti-PF4/heparin Abs were negative by ELISA at the time of the initial thrombocytopenic event, but became positive at the time of the second event. The other patient (No. 4) was given UFH again to prevent embolic events secondary to atrial fibrillation because the therapeutic range of the prothrombin time could not be maintained with warfarin; in this patient, HIT was suspected and anti-PF4/heparin Abs were assessed after the second throm-
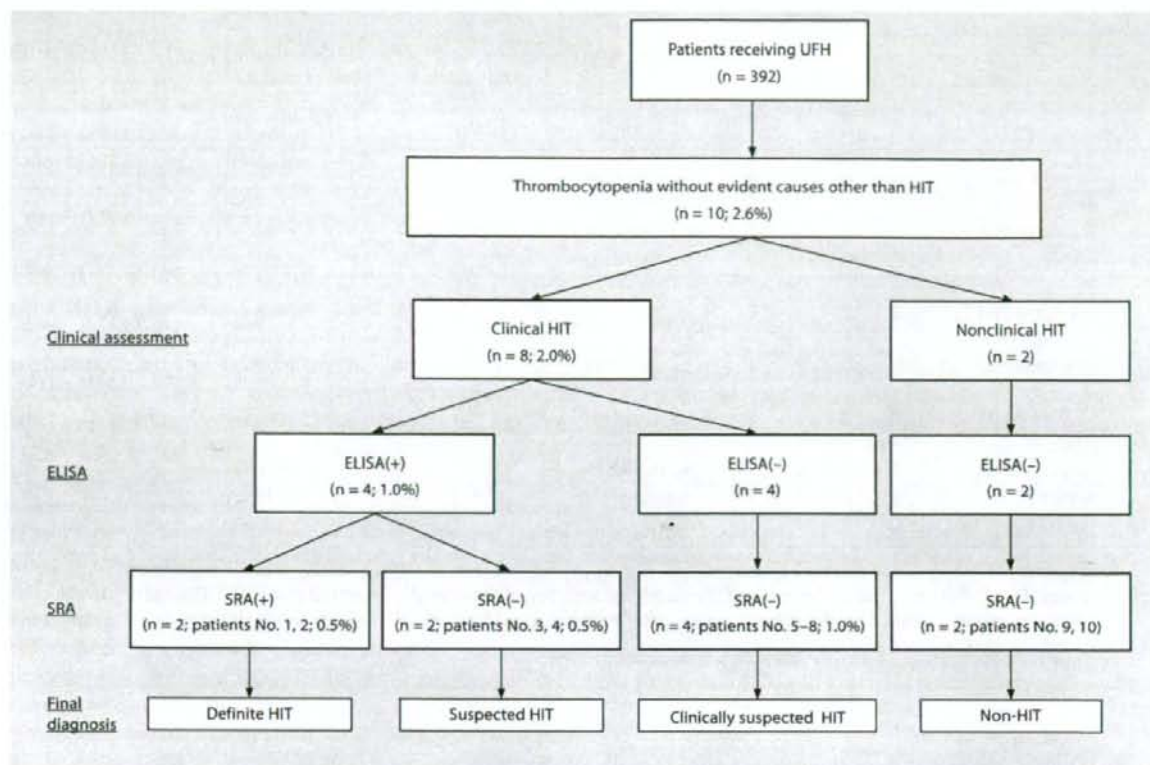
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Fig. 1. Final diagnosis of the patients.

bocytopenic event. Based on these two scoring systems, 8 of 10 patients (No. 1–8) were diagnosed as having clinical HIT. One patient (No. 9) had an intermediate score using the first system (4Ts) and was considered unlikely HIT using the second scoring system; he was later diagnosed as having nonovert disseminated intravascular coagulation. The last patient (No. 10) who did not meet the criteria of clinical HIT in either system was later diagnosed as having idiopathic thrombocytopenic purpura. Anti-PF4/heparin Abs were positive by ELISA in 4 patients (No. 1–4), and by SRA in 2 patients (No. 1, 2). Thus, of 392 stroke patients treated with UFH, patients with definite HIT (No. 1, 2) accounted for 0.5%, patients with suspected HIT (No. 3, 4) accounted for 0.5%, and those with clinically suspected HIT (No. 5–8) accounted for 1.0% (fig. 1). The optical densities of anti-PF4/heparin Abs by ELISA were similar in patients with definite HIT and patients with suspected HIT (table 1).

The 8 patients who were diagnosed as having clinical HIT had one or more vascular risk factors (table 2). Seven patients (except for No. 6) were diagnosed as having cardioembolic stroke due to atrial fibrillation, prosthetic mitral valve, nonbacterial thrombotic endocarditis (identified by autopsy examination), and left ventricular dyskinesis. The remaining patient (No. 6) was thought to have had an embolic stroke, even though no embolic source were identified. In all patients, culprit infarcts were located in the carotid arterial territory; 1 patient also had bilateral cerebellar infarcts (No. 5). The admission NIHSS score varied between 7 and 18 (median 16.5) in all patients, and was higher than that of the other 384 stroke patients treated with UFH (interquartile range 3–16, median 7, p = 0.033).

In these 8 patients, the total dose of UFH given before the nadir of the platelet count varied from 10 to 375 (median 95) $\times$ 10^3 units (principally, 10×10^3 units/day

The doses were greater in patients with definite and suspected HIT than in those with clinically suspected HIT (p = 0.043). The platelet count dropped to its nadir between 2 and 37 (median 9) days after the initiation of UFH, and blood samples for the assays were collected between 5 and 28 (median 9) days after the initiation of UFH (fig. 2; table 2). The interval between the day of platelet count nadir and that of blood collection was within 2 days for all but 1 patient (No. 3). It should be noted that the thrombocytopenia in the definite HIT patients (No. 1, 2) occurred between days 5 and 10 of heparin therapy, which is the highest risk window for HIT, with prompt recovery of platelet count after the cessation of UFH and the administration of argatroban as described below. The platelet count reached its nadir at 20 days or later in both patients with suspected HIT (No. 3, 4) and at 2 days in 1 of the patients with clinically suspected HIT (No. 6). There were no significant differences between patients with definite or suspected HIT and those with clinically suspected HIT in baseline platelet counts (p = 0.248), nadir platelet counts (p = 0.773), and in the change in nadir/baseline counts (p = 0.564). For all but 1 patient with clinically suspected HIT (No. 8), UFH administration was stopped immediately when HIT was suspected (table 2). For definite and suspected HIT patients, intravenous argatroban was given as an alternative to UFH to maintain the activated partial thromboplastin time within 1.5- and 3.0-fold of the baseline value.

One patient with suspected HIT (No. 3) and 2 patients with clinically suspected HIT (No. 5, 8) developed thrombotic events. Patient No. 3 developed pain in his right leg on the eighth day of acute UFH treatment and was diagnosed as having DVT on ultrasound and PE on lung scintigraphy. Intravenous argatroban was given between days 28 and 41 after the initial stroke onset, but 44 days and 66 days after the initial stroke, this patient had recurrent embolic strokes probably due to nonbacterial thrombotic endocarditis secondary to the lung adenocarcinoma. Sixty days after the initial stroke, he also developed an acute myocardial infarction. These repeated events may have been due to a hypercoagulable state related to the adenocarcinoma [22]. The other 3 patients with definite or suspected HIT who were treated with argatroban did not develop thromboembolic events. Patient No. 5 developed a recurrent cardioembolic stroke 8 days after the initiation of UFH, at which time she was also receiving oral warfarin due to the presence of a prosthetic mitral valve and atrial fibrillation; the international normalized ratio of the prothrombin time was 3.11. Patient No. 8 developed a DVT that was detected on ultrasound when her
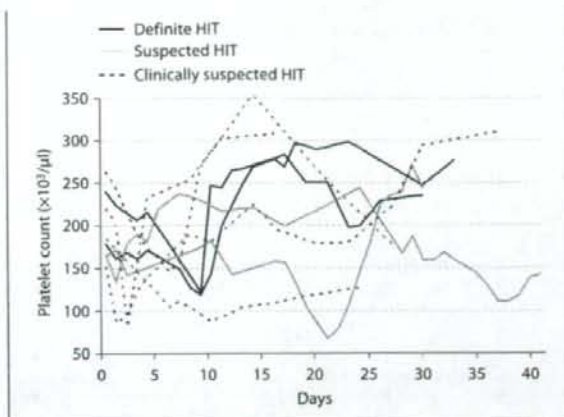

Fig. 2. Serial platelet counts of the patients.

left leg became swollen 8 days after initiation of UFH; UFH therapy was stopped, and she was given elastic stockings.

Both patients with suspected HIT died (No. 3, 4); patient No. 3 died of lung cancer 3 months after the initial stroke event, and patient No. 4 died of acute respiratory distress syndrome 30 days after the stroke event. The remaining 6 patients needed assistance for activities of daily living; their mRS score was 3 or more. The mRS score of the 8 patients was higher than that of the other 384 stroke patients treated with UFH (interquartile range 1–4, median 2, p = 0.001).

Discussion

In this study, we determined the incidence of HIT and clarified the clinical picture of patients who developed HIT during the acute management of ischemic stroke using UFH. The diagnosis of HIT was made both clinically and serologically, in particular using SRA. The first major finding of this study was that 2.0% of the stroke patient population had HIT based on the two clinical scoring systems: 1.0% had both clinical and serological HIT; 0.5% had definite HIT confirmed by SRA. The second major finding was that the clinical severity and outcome of acute stroke patients who were suspected of having HIT were relatively unfavorable compared to other acute stroke patients.

Both ELISA and SRA are similarly and highly sensitive (>95%) for clinical HIT [23–25]. However, the speci-