Delporte and colleagues[24] have recently suggested a causal relationship exists between leptin and adiponectin in OB mice. The adiponectin content of visceral fat and adiponectin secretion by adipose tissue were blunted in OB mice. Leptin treatment of OB mice increased adiponectin mRNA, adiponectin content, and secretion from visceral fat by 50-80%. Leptin also directly stimulated adiponectin mRNA and secretion from adipocytes. Leptin replacement therapy was reported to restore adipose tissue adiponectin concentration and secretion, at least in part, via direct stimulation of adiponectin gene expression in OB mice. Therefore, we believe that leptin treatment might induce expression of cardiac adiponectin mRNA and content in OB mice with viral myocarditis through a similar mechanism.

We determined the expression levels of a proinflammatory cytokine, TNF-α mRNA, and protein in the heart from different mice on days 4 and 8 after viral inoculation as previously described.[8] Significantly increased levels of cardiac TNF-α mRNA and protein on both days were observed in OB mice compared with those in WT mice, together with severe myocarditis and reciprocal changes in adiponectin mRNA and protein. Administration of leptin to OB mice inhibited the progression of severe myocarditis through augmentation of adiponectin mRNA and protein levels and reduced levels of TNF-α mRNA and protein. Adiponectin-knockout mice revealed a high level of TNF-α mRNA in adipose tissue and a high plasma TNF-α concentration.[12] Adiponectin was also reported to inhibit TNF-α production in macrophages.[21] Therefore, adiponectin expression induced by leptin replacement seems to be able to protect against myocarditis through suppression of expression of TNF-α.

Cloning of cDNA encoding two adiponectin receptors (AdipoR1 and AdipoR2) has recently been demonstrated.[25] AdipoR1 is located at chromosome 1p36.13-q41, and AdipoR2 is located at chromosome 12p13.31. AdipoR1 and AdipoR2 are expressed ubiquitously in most organs, especially AdipoR1 in skeletal muscle and AdipoR2 in the liver.[25] Pancreatic β cells were also shown to express adiponectin receptors in a cell culture system.[26] These receptors include 7 transmembrane domains and activate signaling molecules such as PPAR-α, AMP-activated protein kinase, and mitogen-activated protein kinase.[25] Interestingly, we found AdipoR1 and AdipoR2 immunoreactivity as well as adiponectin immunoreactivity in myocardial cells in a mouse model of viral myocarditis. These results indicate that the surviving myocytes may possess an adiponectin-autocrine system, which leads to protection against the progression of myocardial inflammation.

Obesity reduces AdipoR1/R2 expression levels and decreases adiponectin sensitivity, which finally leads to insulin resistance. Improved adiponectin sensitivity should serve as a treatment target for obesity-related diseases.[27] Replace-

ment of leptin in OB mice inhibited the development of severe myocarditis through augmentation of AdipoR1, but not AdipoR2, immunoreactivity in myocytes. We found that adiponectin replacement therapy could attenuate myocardial damage in OB mice with viral myocarditis.[28] Adiponectin replacement also suppressed the development of severe myocarditis by enhancing myocyte AdipoR1, but not AdipoR2, immunoreactivity. Thus, there seems to be selectivity in the adiponectin receptor subtype in myocytes from OB mice. Similarly to our data, it has been demonstrated in another experiment that elevation of liver AdipoR1 expression and reduction in the AdipoR2 expression were observed in obese mice given a combination of leptin and a melanocortin receptor agonist, although there were no alterations in the AdipoR1 or AdipoR2 expression in the mice receiving the melanocortin receptor agonist alone.[29] In addition, changes in AdipoR1/R2 expression in skeletal muscle and adipose tissue have been reported in type 2 diabetic patients during PPAR-γ agonist therapy.[30] AdipoR1 expression was up-regulated in adipose tissue but down-regulated in muscle by rosiglitazone. On the other hand, AdipoR2 expression was not changed by rosiglitazone in either of the tissues. The increase in adipose tissue AdipoR1 expression with rosiglitazone was associated with increased postprandial triglyceride clearance and increased fasting fatty acid output. AdipoR1 is suggested to play a role in mediating adiponectin effects in specific tissues in relation to insulin sensitization and anti-inflammation. It is necessary to evaluate the effects of adiponectin using AdipoR1-knock out mice in the future.

Conclusion: We found reduced expression of adiponectin mRNA, immunoreactivity, and protein levels and decreased immunoreactivity of AdipoR1 in the heart from OB mice after viral infection, together with increased heart weight, severe myocardial inflammation, and elevated levels of cardiac TNF-α mRNA and protein. Administration of leptin to OB mice resulted in suppression of the development of severe myocarditis through augmentation of adiponectin mRNA, immunoreactivity, and protein levels, elevated AdipoR1 immunoreactivity in myocytes, and reduced levels of TNF-α mRNA and protein. Our observations suggest that impaired expression of adiponectin in the heart is associated with the development of viral myocarditis through enhancement of TNF-α expression under a leptin-deficient status.

ACKNOWLEDGMENTS

This study was supported in part by a Grant for Promoted Research from Kanazawa Medical University (S2004-2 and S2005-5), a Grant for Project Research from the High-Technology Center of Kanazawa Medical University (H2004-7), and the Science Research Promotion Fund of the Promotion and Mutual Aid Corporation for Private Schools of Japan.

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Original Article

Angiotensin I–Converting Enzyme Inhibitor Improves Reactive Hyperemia in Elderly Hypertensives with Arteriosclerosis Obliterans

Masashi OKURO[1)], Shigeto MORIMOTO[1)], Takashi TAKAHASHI[2)], Kohya OKAISHI[1)], Takeshi NAKAHASHI[1)], Hiroshi MURAI[1)], Kunimitsu IWAI[1)], Tsugiyasu KANDA[2)], and Masayuki MATSUMOTO[1)]

Endothelial function in elderly hypertensive patients with arteriosclerosis obliterans has not been evaluated. We examined whether antihypertensive drugs improve vasodilatory response to reactive hyperemia of the limbs in elderly hypertensive patients (83±8 [SD] years) without (n=46, 0.9 ≤ ankle-brachial pressure index ≤1.4) and with (n=24) arteriosclerosis obliterans (ankle-brachial pressure index <0.2). Patients were randomized for treatment with monotherapy of either temocapril (14 with and 26 without arteriosclerosis obliterans) or amlodipine (10 with and 20 without arteriosclerosis obliterans) for 6 months. Blood flows of the forearms and legs were measured by strain-gauge plethysmography. The vasodilatory response to the release of compression of the forearms and thighs at 200 mmHg or 20 mmHg more than systolic blood pressure for 5 min and to sublingual administration of nitroglycerin (0.3 mg) was assessed. The maximum reactive hyperemic flow in 35 legs with arteriosclerosis obliterans was significantly (p<0.001) decreased compared to the value in legs in the control hypertensive subjects. Moreover, maximum reactive hyperemic flow in the forearms of patients with arteriosclerosis obliterans was significantly (p=0.002) decreased compared to that in the control subjects. Blood pressure was similarly decreased by treatment with temocapril or amlodipine. Response to nitroglycerin (0.3 mg) was not changed by either drug. Treatment with temocapril significantly improved maximum reactive hyperemic flow of not only the legs and forearms in control hypertensives but also the legs and forearms in patients with arteriosclerosis obliterans, and attenuated the worsening of activity of daily living in these patients, although treatment with amlodipine did not. These results suggest that the angiotensin-converting enzyme inhibitor temocapril has a beneficial effect on endothelial function in elderly patients with arteriosclerosis obliterans. (*Hypertens Res* 2006; 29: 655–663)

Key Words: antihypertensive drug, arteriosclerosis obliterans, elderly, hypertension, plethysmography

Introduction

Arteriosclerosis obliterans has become one of the major health problems in the elderly, since the incidence of this disease increases with age (*1*), and since the disease not only causes decreased activity of daily living and quality of life, but is also a serious life-threatening condition due to its progressive nature and/or the high mortality of associated arterial lesions of cardiovascular organs (*2*). Arteriosclerosis obliterans frequently follows an inexorable downhill course (*3*, *4*), and, importantly, the Consensus Document of the European

From the Departments of [1)]Geriatric Medicine and [2)]General Medicine, Kanazawa Medical University, Ishikawa, Japan.

This study was supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Health, Labour and Welfare of Japan (to S.M.) and a grant for promoted research from Kanazawa Medical University (S2005-5 to T.T.).

Address for Reprints: Shigeto Morimoto, M.D., Ph.D., Department of Geriatric Medicine, Kanazawa Medical University, 1–1 Daigaku, Uchinada-machi, Kahoku-gun, Ishikawa 920–0293, Japan. E-mail: shigeto@kanazawa-med.ac.jp

Received January 24, 2006; Accepted in revised form May 29, 2006.

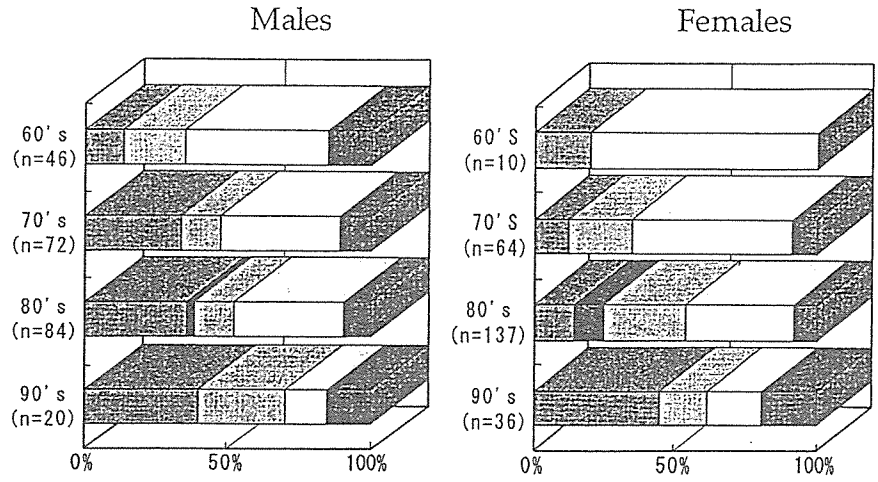

Fig. 1. *Prevalence of subjects with ABI>1.4 (■), 0.9≤ABI≤1.4 (□), 0.5≤ABI<0.9 (▨), 0.2≤ABI<0.5 (■), and ABI<0.2 (■) in males (n=222) and females (n=247) in their 60s, 70s, 80s and 90s respectively, in a hospital for the elderly.*

Working Group on Critical Limb Ischemia has concluded that there is no optimal medical therapy for this disease (*5*).

On the other hand, arteriosclerosis obliterans is highly associated with hypertension (*6*), and hypertension itself is a major risk for arteriosclerosis obliterans (*7–9*). It has been shown by microscopic examination that the endothelium of lesions of arteriosclerosis obliterans is impaired (*10*), and that endothelial function assessed by maximum reactive hyperemic flow is significantly decreased in patients with arteriosclerosis obliterans and/or hypertension (*11*). Among antihypertensive agents, angiotensin I–converting enzyme (ACE) inhibitors (*12, 13*) and calcium antagonists (*13, 14*) are known to maintain or even increase limb blood flow and to improve tolerability to exercise in patients with arteriosclerosis obliterans. Moreover, ACE inhibitors have been reported to have beneficial effects on endothelial function, as assessed by reactive hyperemia in middle-aged patients with essential hypertension (*15*) and in elderly patients with hypertension (*16*). However, there has been little study of the effects of these antihypertensive agents on endothelial function in this hypertension-associated disease.

In this study, we evaluated the effects of an ACE inhibitor, temocapril, and a calcium antagonist, amlodipine, on vasodilator responses in the limbs of elderly patients with hypertension with or without associated arteriosclerosis obliterans. These drugs were given as monotherapy for 6 months. We examined whether reactive hyperemia was impaired, and whether the antihypertensive agents affected the vasodilatory responses in these elderly subjects.

Methods

Study Population

The study was conducted in Sengi Hospital, which serves as both a hospital and a long-term care facility for the elderly; medical and care services are often combined in single facilities in Japan (*17*). Prior to the selection of subjects, we surveyed ankle-brachial pressure index (ABI) in 469 elderly inpatients aged 65 years or older (222 males and 247 females, including 46 males and 10 females in their 60s, 72 males and 64 females in their 70s, 84 males and 127 females in their 80s, and 20 males and 36 females in their 90s, with a mean [±SD] age of 82±9 years) in the hospital. ABI was measured as described below (*18*). Our classification of ABI was essentially based on the report of Resnick *et al.* (*18*), who used the three categories of ABI>1.4 (at least one leg), 0.9≤ABI≤1.4 (both legs), and ABI<0.9 (at least one leg). However, since about half (48%) of our subjects had an ABI<0.9, we further separated the category of ABI<0.9 into three classes, namely, 0.5≤ABI<0.9, 0.2≤ABI<0.5, and ABI<0.2 (at least one leg, respectively). The preliminary survey revealed that the numbers (%) of subjects with ABI>1.4, 0.9≤ABI≤1.4, 0.5≤ABI<0.9, 0.2≤ABI<0.5, and ABI<0.2 were 51 (12%), 191 (41%), 97 (21%), 16 (3%), and 114 (24%), respectively. Increases in age-related prevalence in those with ABI<0.2 were prominent both in males and females (Fig. 1). Moreover, the survey also disclosed that the percentage of subjects with 0.5≤ABI<0.9 was very small. Furthermore, none of the 113 subjects with 0.2≤ABI<0.9 showed any characteristic features of arteriosclerosis obliterans, such as intermittent claudication, pain at rest, or ulcer/gangrene of the lower extremities, although 12 (11%) of the 114 subjects with ABI<0.2 showed characteristic features of arteriosclerosis obliterans, *i.e.*, pain at rest (8 patients) and ulcer/gangrene (4 patients). Accordingly, we adopted rather strict criteria, using ABI<0.2 for selection of hypertensive patients with arteriosclerosis obliterans. The numbers (%) of subjects with hypertension, defined as a systolic blood pressure (SBP) of 140 mmHg or higher and/or diastolic blood pressure (DBP) of 90

mmHg or higher in the sitting position on at least three separate occasions and/or current use of antihypertensive drugs, were 19 (37%) out of 51 subjects with ABI>1.4, 67 (35%) out of the 191 with 0.9≤ABI≤1.4, 35 (36%) out of the 97 with 0.5≤ABI<0.9, 5 (31%) out of the 16 with 0.2≤ABI<0.5, and 57 (50%) out of the 114 with ABI<0.2. From among these hypertensive patients, we selected elderly patients with mild (140–159/90–99 mmHg) to moderate (160–179/100–109 mmHg) hypertension (mean±SD age: 83±8 years; age range: 68–94 years) without (n=46, 0.9≤ABI≤1.4) or with (n=24) arteriosclerosis obliterans (ABI<0.2, two legs in 11 subjects and one leg in 13 patients). Subjects showing pain at rest or ulcer/gangrene as features of arteriosclerosis obliterans, or those with diabetes mellitus (fasting blood glucose >7 mmol/l or treated with or current use of an antidiabetic drug) or hypercholesterolemia (total cholesterol >5.66 mmol/l) were excluded by clinical and laboratory examinations. None of the subjects had undergone amputation due to arteriosclerosis obliterans. Patients were randomized to treatment with initial doses of 2 mg temocapril (14 patients with and 26 patients without arteriosclerosis obliterans; Sankyo Co., Ltd., Tokyo, Japan) or 2.5 mg amlodipine (10 patients with and 20 patients without arteriosclerosis obliterans; Sumitomo Pharmaceutical Ltd., Osaka, Japan) once daily for 6 months. When the blood pressure was not decreased by 20/10 mmHg or to below 150/90 mmHg, the dose of drug was doubled. Blood pressure control was achieved in all hypertensive subjects with monotherapy of either drug. All patients had either never been treated (n=35) or had discontinued antihypertensive drugs—in this case, β-blockers (n=2), ACE inhibitors (n=17), or calcium antagonists (n=16)—for at least 4 weeks before the study. Biochemical factors were measured in blood collected in the morning after overnight fasting. The study protocol was approved by the ethical committee of the hospital. All subjects were Japanese, and only those who gave informed consent were enrolled in the study.

Measurement of Ankle and Arm Blood Pressures

At each ABI measurement, right arm blood pressures and bilateral ankle blood pressure (posterior tibial artery), measured by handheld Doppler ultrasonography (Imex Medical Systems, Golden, USA), were taken with the subject supine. If the absent pulse was verified, ankle blood pressure measures were taken on the dorsalis pedis. The means of the 2 measurements for each leg and for the arm were used to calculate ABI, and the lower of the 2 values was used to define ABI for each individual (*18*).

Blood Flow Measurements of the Forearm and Legs

The study was conducted in a dark, quiet, temperature-controlled (at 23°C) room. Subjects rested for 30 min in the supine position before the study. Blood flow of the right forearm and of both legs was measured by strain gauge plethysmography (model EC6; De Hokanson, Inc., Bellevue, USA) (*19*, *20*). Hand and foot circulations were excluded by wrist and ankle cuffs inflated to suprasystolic pressure. Mercury-in-silastic strain gauges that had been electrically calibrated were placed on the widest part of the right forearm and both lower legs at 5 cm and 8 cm below the antecubital and popliteal creases, respectively. Blood flows of forearm or legs were calculated from the rate of increase in forearm or leg volumes, while venous return was prevented by inflating the cuffs at upper arm or thigh to a venous-occlusion pressure of 50 mmHg. Flow measurements were recorded for 9 s every 15 s, and an average of 4 measurements was used for analysis. Endothelium-dependent vasodilatation was assessed by ischemia-induced reactive hyperemia. After a baseline recording of 4 min, ischemia was induced for 5 min by inflating the upper arm cuff to 200 mmHg or 20 mmHg more than SBP. Immediately after cuff deflation, maximal hyperemic blood flow was measured (peak flow), followed by continuous measurements for 3 min. At least 15 min after the last measurement, blood flow was measured to confirm that it had returned to the basal level. Then, endothelium-independent vasorelaxation was assessed after sublingual administration of nitroglycerin at 0.3 mg by one puff of a spray device (Miokol Spray; Toa Eiyo, Tokyo, Japan), and blood flows were measured for 5 min. The blood flow is expressed as ml of blood per min per 100 ml of limb volume (*15*, *20*, *21*).

Other Measurements

We evaluated activity of daily living according to the following four states of ambulation: walking, using a wheelchair, sitting on the bed, and bedridden. We also recorded known risk factors for arteriosclerosis obliterans, including dementia (Mini-Mental State Examination score≤23), chronic stage of stroke (motor deficit and evidence of cerebral hemispheric stroke on CT or MRI), and chronic ischemic heart disease (previous myocardial infarction or angina pectoris).

Statistical Analyses

Data are expressed as the mean±SD. Differences in changes of blood pressure and vasodilatory responses by treatment with antihypertensive drugs were assessed by analysis of variance with repeated measurements. Changes in ABI and activity of daily living were assessed by non-parametric Wilcoxon test. A value of $p<0.05$ was regarded as significant. Differences among the four groups were analyzed by Kruskal-Wallis χ^2 and Mann-Whitney U test for multiple comparisons with post-hoc Bonferroni correction. The statistical significance of p values was set at 0.008 for the analysis among four groups. Data were analyzed on a microcomputer running SPSS (SPSS Inc., Chicago, USA).

Table 1. Comparison of Clinical Factors between Hypertensive Patients with and without Arteriosclerosis Obliterans

	Control subjects		Arteriosclerotic patients	
	Amlodipine (n=20)	Temocapril (n=26)	Amlodipine (n=10)	Temocapril (n=14)
Clinical background				
Age (years)	82±7	83±8	83±7	83±8
Sex (male/female)	7/13	10/16	4/6	5/9
Total cholesterol (mmol/l)	4.89±0.32	4.86±0.30	4.85±0.29	4.87±0.28
Fasting blood glucose (mmol/l)	5.06±0.34	5.03±0.20	5.04±0.31	5.04±0.29
Dementia (n (%))	13 (65)	16 (62)	6 (60)	8 (57)
Chronic stage of stroke (n (%))	4 (20)	8 (31)	2 (20)	3 (21)
Ischemic heart disease (n (%))	7 (35)	8 (31)	3 (30)	3 (21)
Before				
SBP (mmHg)	161±12	162±13	162±14	162±12
DBP (mmHg)	82±7	84±8	83±7	82±6
Heart rate (bpm)	68±5	69±7	67±6	68±7
ABI	1.03±0.09	1.04±0.10	0.11±0.010#	0.10±0.01#
Activity of daily living (n (%))				
Walking	11 (55)	15 (57)	6 (60)	8 (57)
Using a wheelchair	5 (25)	6 (23)	2 (20)	4 (29)
Sitting on the bed	2 (10)	2 (8)	1 (10)	1 (7)
Bedridden	2 (15)	3 (12)	1 (10)	1 (7)
Six months				
SBP (mmHg)	138±10†	137±11†	138±10†	139±12†
DBP (mmHg)	72±7†	74±8†	72±9†	74±8†
Heart rate (bpm)	69±5	71±7	69±6	69±8
ABI	0.99±0.05	1.05±0.03	0.10±0.01#	0.13±0.01#,†
Activity of daily living (n (%))				
Walking	10 (50)	15 (57)	4 (40)	10 (72)
Using a wheelchair	3 (15)	5 (19)	1 (10)	2 (14)
Sitting on the bed	4 (20)	3 (12)	2 (20)	0 (0)
Bedridden	3 (15)	3 (12)	3 (30)	2 (14)

SBP, systolic blood pressure; DBP, diastolic blood pressure; bpm, beats per minute; ABI, ankle-brachial pressure index. Values are mean±SD. #$p<0.008$: significant difference *vs.* control groups by Kruskal-Wallis analysis. *$p<0.05$ and †$p<0.01$: significant difference *vs.* before.

Results

Table 1 shows the backgrounds of the groups of hypertensive elderly patients with and without arteriosclerosis obliterans treated with amlodipine or temocapril. There was no statistically significant difference among the four groups in age, male/female ratio, circulating concentration of total cholesterol or glucose, activity of daily living, or prevalence of dementia, chronic phase of stroke, or chronic ischemic heart disease. Of course, the ABI values in the two groups with arteriosclerosis obliterans were significantly ($p<0.008$) reduced compared to those in the two control groups (Table 1).

The values of basal blood flow (Fig. 2a), nitroglycerin-induced increase in blood flow (Fig. 2b), and maximum reactive hyperemic flow (Fig. 2c) in the 35 affected legs of the 24 hypertensive patients with arteriosclerosis obliterans were significantly ($p<0.001$ in each) decreased compared to the respective values in the 92 legs of 46 age- and sex-matched elderly hypertensive control subjects. Although the basal and nitroglycerin-induced increases in blood flow in the right forearm were similar in the two hypertensive groups, maximum reactive hyperemic flow in the right forearm in the 24 patients with arteriosclerosis obliterans was significantly ($p<0.01$) decreased compared to that in the 46 control subjects.

Baseline values (ml/min/100 ml) of blood flow in the legs and forearms before treatment in patients with arteriosclerosis obliterans (1.8±0.4 and 3.9±0.5 in the amlodipine-treated group and 1.9±0.5 and 4.0±0.4 in the temocapril-treated group) and patients without arteriosclerosis obliterans (3.2±0.4 and 4.1±0.4 in the amlodipine-treated group and 3.3±0.4 and 4.2±0.5 in the temocapril-treated group) were

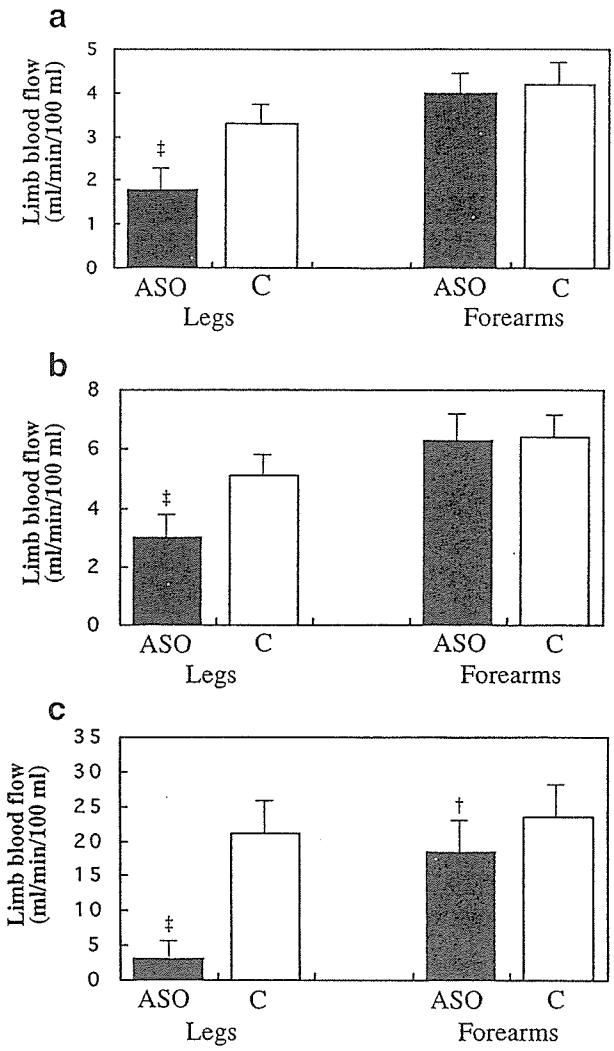

Fig. 2. *Comparison of basal blood flow (a), nitroglycerin-induced increase in blood flow (b), and maximum reactive hyperemic flow (c) between legs with arteriosclerosis obliterans (ASO, n=35, solid bars) and legs in control hypertensive patients (C, n=92, open bars) (left), and between the right forearms in ASO patients (n=24) and control patients (n=46) (right).* †$p<0.01$, ‡$p<0.001$: *significant differences between hypertensive elderly patients with and without ASO.*

similar in the two drug groups. Basal blood flow in the affected legs of patients with arteriosclerosis obliterans ($n=14$ in the amlodipine-treated group and $n=21$ in the temocapril group) was again significantly lower compared to that in the legs of patients without arteriosclerosis obliterans in the respective drug groups.

Blood pressure was well controlled by 2.5 mg ($n=6$ in the group with arteriosclerosis obliterans and $n=12$ in the control group) or 5 mg ($n=4$ in the group with arteriosclerosis obliterans and $n=8$ in the control group) amlodipine, or 2 mg ($n=11$ in the group with arteriosclerosis obliterans and $n=22$ in the control group) or 4 mg ($n=3$ in the group with arteriosclerosis obliterans and $n=5$ in the control group) temocapril. Monotherapy of either amlodipine or temocapril for 6 months produced a similar reduction of blood pressure (Table 1).

Treatment with temocapril slightly but significantly increased the ABI value in patients with arteriosclerosis obliterans, although amlodipine did not significantly change the ABI value in patients with arteriosclerosis obliterans (Table 1). On the other hand, basal blood flow and nitroglycerin-induced increase in blood flow (Fig. 3a) in the legs and forearm were not significantly changed by either drug. Treatment with temocapril significantly improved maximum reactive hyperemic flow (ml/min/100 ml) of not only the legs (21.8 ± 3.8 *vs.* 24.9 ± 3.7, $p<0.001$) and forearm (24.2 ± 3.6 *vs.* 27.3 ± 3.6, $p<0.01$) in control hypertensive subjects, but also in the affected legs (2.8 ± 2.2 *vs.* 5.8 ± 2.5, $p<0.001$) and forearms (18.2 ± 3.8 *vs.* 22.3 ± 2.9, $p<0.01$) of patients with arteriosclerosis obliterans, although treatment with amlodipine did not affect maximum reactive hyperemic flow (Fig. 3b). Moreover, the activities of daily living were slightly but significantly decreased in control subjects and patients with arteriosclerosis obliterans treated with amlodipine for 6 months, but not in the other two groups treated with temocapril (Table 1).

Discussion

As a diagnostic criterion for arteriosclerosis obliterans, ABI<0.9 is often used (*18, 22*). According to one report, the prevalence of asymptomatic arteriosclerosis obliterans defined by an ABI value <0.9 was only 3.4% in subjects aged 65 years or more who were inhabitants of rural communities in Japan (*22*). However, our preliminary survey in 469 inpatients of an elderly hospital aged 65 years or older revealed that about half of the elderly subjects would have been diagnosed with arteriosclerosis obliterans if this criterion had been adopted. Moreover, subjects with $0.2\leq$ABI<0.9 did not show any characteristic features of arteriosclerosis obliterans. Furthermore, only 3% of the total subject group had $0.2\leq$ABI<0.5. According to these findings, we selected subjects with ABI<0.2 as subjects with arteriosclerosis obliterans in this study. Furthermore, subjects with apparent rest pain or ulcer/gangrene were excluded from the present study, and thus most of the remaining subjects would be classified as stage I or II in Fontaine classification. However, we did not adopt the Fontaine classification for staging of the severity of arteriosclerosis obliterans, since many of the inpatients of the elderly hospital were not capable of complaining about intermittent claudication or rest pain of the lower extremities, and in some cases not capable of walking, due to rather high prevalence of dementia and chronic phase of stroke (Table 1). Instead, we analyzed the activity of daily living in the four groups, and found no statistically significant difference in this parameter among the four groups. This observation suggests that decreased activity of daily living due to arteriosclerosis

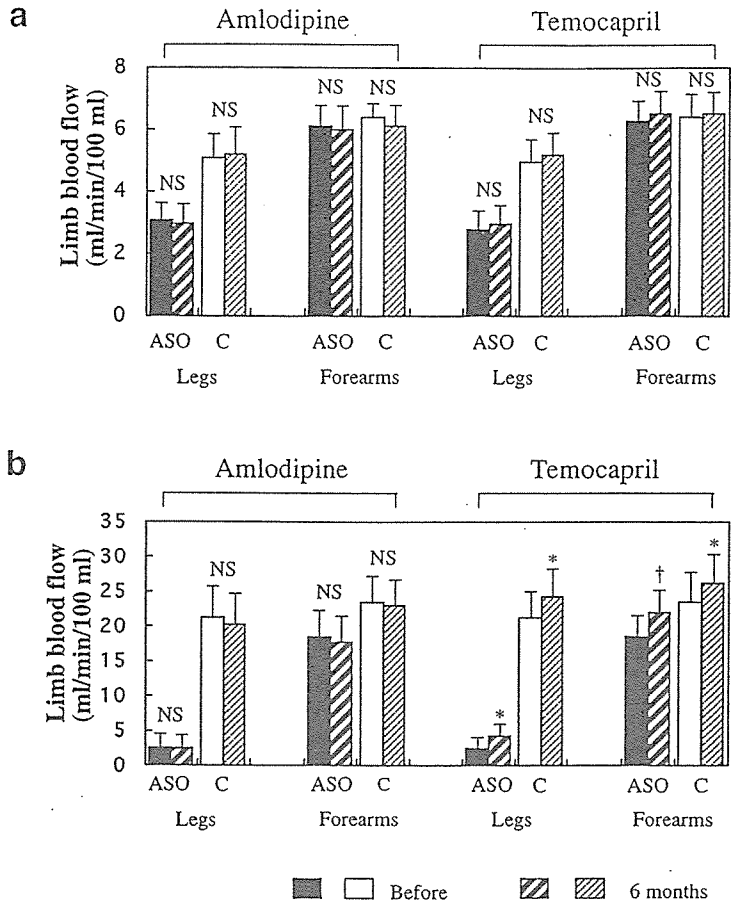

Fig. 3. *Changes of blood flow in legs and forearms in response to sublingual administration of nitroglycerin (a), and of reactive hyperemic response (b), before and after 6 months of treatment with amlodipine or temocapril. ASO, patients with arteriosclerosis obliterans; C, control patients.* *$p<0.05$, †$p<0.01$: *significant differences between before and after 6 months of treatment; NS: not significant.*

obliterans itself was not a direct cause of the differences seen in the basal and reactive hyperemic blood flow in this study.

There may be two possible mechanisms which could explain the decreased reactive hyperemia in the diseased legs. Since we measured ankle blood pressure at the lower part of the main coarctation sites of arteriosclerosis obliterans (*23*), the first possibility is that the endothelial function had already reached an almost maximal level because of the decreased blood stream due to coactation of the upper part of the legs which would have resulted in a decrease in reactive hyperemia after complete occlusion by the cuff. The second possibility is that the decreased reactive hyperemia in the diseased legs was one of the general findings of the reduced vasodilatory endothelial function represented by those in upper extremities, since vasodilatory responses to reactive hyperemia were significantly decreased not only in the diseased legs but also in the forearms of patients with arteriosclerosis obliterans. The results of several reports may lend support to this latter mechanism. Postischemic hyperemia in human limbs is thought to be partly mediated by nitric oxide (NO), since inhibition of NO synthesis by N^G-monomethyl-L-arginine has been shown to decrease reactive hyperemic flow (*24*), although the participation of other mechanisms, including endothelium-dependent vasodilating substances such as prostaglandins (*25*) and endothelium-stimulating substances such as adenosine (*26*), has also been reported. Impaired endothelial function in hypertensive patients has been demonstrated by several investigators (*15*, *27–29*). Moreover, aging itself is also a causal factor for decreased endothelial function (*30*). Furthermore, a generalized decrease of endothelial function was also reported in patients with thromboangiitis obliterans (Buerger's disease), another occlusive arterial disease (*31*). In the present study, on the other hand, treatment with temocapril slightly but significantly increased the ABI value, and improved the reactive hyperthermia even in the legs with arteriosclerosis obliterans without any significant change in basal blood flow, suggesting that this ACE inhibitor improves not only the generalized decrease in endothelial function in patients with arteriosclerosis obliterans, but also might improve the decreased reactivity of the endothelial function

due to defatigation by continuous stimulation of the decreased blood stream.

Our results also showed that the reactive hyperemia in the legs and forearms in control hypertensive subjects as well as in the affected legs and forearms of patients with arteriosclerosis obliterans was improved by 6 months of treatment with an ACE inhibitor, temocapril, but not by a calcium antagonist, amlodipine. Moreover, significant decreases in the activity of daily living observed both in control subjects and patients with arteriosclerosis obliterans treated with amlodipine for 6 months were apparently attenuated by treatment with the ACE inhibitor. Treatment with amlodipine and temocapril produced a similar blood pressure reduction after 6 months of treatment. Both drugs are long-acting antihypertensive agents that are usually given once a day. Therefore, it is unlikely that the difference in duration of normotension can explain the results obtained in this study. ACE inhibitors reduce circulating and tissue levels of angiotensin II (*32*), the most potent vasoconstrictor, which causes sequential production of other vasoconstricting factors such as endothelin (*33*) and prostaglandin H_2 (*34*) from endothelial cells, and production of superoxide, an inactivator of NO, *via* the stimulation of reduced nicotine amide adenine dinucleotide oxidase (*35*). Moreover, ACE inhibitors induce accumulation of bradykinin (*36*), which causes the release of endothelium-derived relaxing factor from the endothelium (*37*) and the ACE inhibitor–induced increase in reactive hyperemia (*38*). These mechanisms may participate in the improvement of vasodilatory responses to reactive hyperemia in temocapril-treated elderly patients with hypertension. Interestingly, it has also recently been reported that brachial flow-mediated vasodilation is significantly correlated with coronary endothelial function and fibrinolytic activity in response to bradykinin in 14 diabetic and 63 non-diabetic subjects (*39*). The results of our study are partly compatible with previous reports demonstrating the beneficial effects of ACE inhibitors on endothelial function as assessed by reactive hyperemia in middle-aged patients with essential hypertension (*15*) and in elderly patients with hypertension (*16*). Moreover, treatment with temocapril might improve the decreased reactivity of endothelial function due to defatigation by continuous stimulation of the decreased blood stream.

In the present study, on the other hand, treatment with amlodipine had no effects on reactive hyperemia in the legs and forearms of elderly hypertensive patients with and without arteriosclerosis obliterans. Since amlodipine (*40*) and other calcium antagonists (*41*) are reported to enhance *in vitro* endothelial synthesis of NO, the reason for the difference in the vasodilatory response between the two antihypertensives is uncertain. However, our results are partly compatible with the clinical observations that treatment with amlodipine failed to improve forearm reactive hyperemia (*15*) and L-arginine–induced increase in renal plasma flow (*42*) in patients with essential hypertension.

In the present study, however, we did not measure factors related to the renin-angiotensin system or bradykinin. Further studies including evaluations of local or systemic levels of these factors are required to elucidate the precise mechanism of the efficacy of ACE inhibitors for improvement of vasodilatory responses to reactive hyperemia, especially in elderly hypertensive patients with arteriosclerosis obliterans, since ACE inhibitors suppress local and systemic formation of angiotensin II (*32*) and degradation of bradykinin (*36*) and since local bradykinin has been reported to play a role in the ACE inhibitor–induced improvement of endothelial function in humans (*38*). Moreover, because they were asymptomatic, despite their rather low ABI values, the majority of elderly inpatients with arteriosclerosis obliterans in this study were not treated with anti-platelet drugs, anticoagulants, or prostaglandins, which are standard prescriptions for younger patients with this disease. The efficacies of these drugs for prevention of progression and treatment of arteriosclerosis obliterans in oldest–old patients like our subjects have not been examined.

Reactive hyperemia-induced increase in forearm blood flow is a frequently used marker of endothelium-dependent vasorelaxation, especially because its measurement is noninvasive (*43*, *44*). However, it has certain limitations compared with the reference method of forearm blood flow measurement during intra-arterial infusion of acetylcholine. Hyperemic blood flow is not exclusively dependent on the endothelium, because in addition to endothelium-derived vasoactive agents, other local metabolic factors may contribute to vasodilatation after ischemia. Furthermore, the placement of the arm occlusion (upper *vs.* lower arm) and the age of the investigated subjects may influence the correlation of hyperemic forearm blood flow with endothelium-mediated vasorelaxation.

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Original Article

Tilting-Induced Decrease in Systolic Blood Pressure in Bedridden Hypertensive Elderly Inpatients: Effects of Azelnidipine

Shigeto MORIMOTO[1], Takashi TAKAHASHI[2], Kohya OKAISHI[3], Takeshi NAKAHASHI[1], Kohji NOMURA[1], Tsugiyasu KANDA[2], Masashi OKURO[1], Hiroshi MURAI[1], Tomoichi NISHINO[3], and Masayuki MATSUMOTO[1]

The object of this study was to examine blood pressure (BP) variability due to postural change in elderly hypertensive patients. The subjects studied were 154 elderly inpatients in a hospital for the elderly (48 male and 106 female; median age: 82 years), consisting of age- and sex-matched bedridden (n=39) and non-bedridden (n=39) normotensive controls and bedridden (n=38) and non-bedridden (n=38) hypertensive patients. BP and pulse rate (PR) were measured in the supine position, then again after a 2-min, 45 deg head-up tilt with the legs horizontal. The decrease in systolic BP (SBP) on tilting in the bedridden hypertensive group (median: −10 mmHg; range: −32 to 9 mmHg) was significantly (p<0.008) greater than those in the other three groups. Monotherapy with azelnidipine, a long-acting calcium channel blocker, for 3 months not only significantly reduced the basal BP and PR of hypertensive patients in the two groups, but also significantly (p<0.05) attenuated the tilt-induced decrease in the SBP to −3 mmHg (−19 to 25 mmHg) and enhanced the change in PR from −1 bpm (−10 to 7 bpm) to 1 bpm (−4 to 23 bpm) in the bedridden hypertensive group. Our findings indicate that tilt-induced decrease in SBP is a rather common phenomenon in bedridden elderly hypertensive patients, and that treatment with azelnidipine attenuates tilt-induced decrease in SBP, probably through an improvement of baroreceptor sensitivity. (*Hypertens Res* 2006; 29: 943–949)

Key Words: **bedridden, head-up tilt, systolic blood pressure, hypertensive elderly, azelnidipine**

Introduction

Increased blood pressure (BP) variability on postural change is a recognized feature in the elderly, especially in those with hypertension (*1*). Moreover, many longitudinal epidemiological studies have shown that increased BP variability on postural change is associated with future cardiovascular events, including coronary heart disease (*2*), stroke (*3*), and even mortality (*4*), and is also recognized as a risk factor for cognitive impairment (*5*) and silent cerebrovascular disease (*6*). In addition, it has recently been reported that lying in a prone posture can lead to unregulated postural hypotension, which has the possibility of being a novel predictor of cardiovascular disease (*7*).

On the other hand, tilting-up of the upper body with the legs horizontal is a commonly performed maneuver in bedridden elderly subjects, and may prevent aspiration pneumonia (*8*).

From the [1]Departments of Geriatric Medicine and [2]General Medicine, Kanazawa Medical University, Ishikawa, Japan; and [3]Department of Medicine, Sengi-Hospital, Ishikawa, Japan.

This study was supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Health, Labour and Welfare of Japan (to S.M.) and a grant for promoted research from Kanazawa Medical University (S2005-5 to T.T.).

Address for Reprints: Shigeto Morimoto, M.D. Ph.D., Department of Geriatric Medicine, Kanazawa Medical University, 1–1 Daigaku, Uchinada-cho, Kahoku-gun, Ishikawa 920–0293, Japan. E-mail: shigeto@kanazawa-med.ac.jp

Received May 2, 2006; Accepted in revised form August 9, 2006.

Table 1. Comparison of Clinical Factors among Normotensive and Hypertensive Inpatients with Non-Bedridden and Bedridden States in a Geriatric Hospital

	NB elderly (n=77)		B elderly (n=77)	
	N (n=39)	H (n=38)	N (n=39)	H (n=38)
Clinical background				
Age (years)	83 (72–93)	82 (70–93)	83 (71–91)	82 (70–92)
Male/female	12/27	12/26	12/27	12/26
Chronic conditions (n (%))				
Dementia	12 (30.8)	11 (28.9)	29 (74.4)$^{\dagger,\#}$	32 (84.2)$^{\dagger,\#}$
Stroke	7 (17.9)	9 (23.7)	29 (74.4)$^{\dagger,\#}$	22 (57.4)$^{\dagger,\#}$
Ischemic heart disease	4 (10.3)	11 (28.9)	8 (20.5)	9 (23.7)
Congestive heart failure	4 (10.3)	4 (10.5)	6 (15.4)	2 (5.3)
Hypoalbuminemia	5 (12.8)	0 (0.0)	11 (28.2)$^{\#}$	6 (15.8)
Diabetes mellitus	0 (0.0)	3 (7.9)	2 (5.1)	0 (0.0)

Values are expressed as median (range) in age. N, normotensives; H, hypertensives; NB, non-bedridden; B, bedridden. $^{\dagger}p<0.008$, *vs.* NB-N group. $^{\#}p<0.008$, *vs.* NB-H group.

However, little has been reported about the BP variability in completely bedridden elderly inpatients. In the present study, we compared variability of BP and pulse rate (PR) at the time of tilting-up of the upper body in non-bedridden and bedridden inpatients with and without hypertension in a hospital for the elderly. We also examined the effects of azelnidipine, a newly developed dihydropyridine-type calcium antagonist that acts without augmentation of the sympathetic nervous system (*9*, *10*), on variability of BP and PR in these hypertensive elderly inpatients.

Methods

Study Subjects

The study was conducted in Sengi-Hospital, a geriatric hospital serving as both a hospital and a long-term care facility for the elderly, which is a common combination of medical and care services in Japan (*11*). Katz's activities of daily living (ADL: bathing, dressing, going to the toilet, transfer, continence, feeding) (*12*) and the Braden scale (*13*) were assessed once a month in all inpatients in the hospital. The research protocol was approved by the Ethics Committee of the hospital. Patients aged 70 years and older, all of whom were Japanese with an admission period of 16 weeks or longer, were invited to participate in the study. All residents who gave informed consent (or whose family members gave consent) were enrolled. Both normotensive and hypertensive subjects were selected from bedridden residents. Control normotensive and hypertensive subjects were age- and sex-matched random samples of non-bedridden subjects admitted to the same hospital. The computerized admission lists served as the sampling frame, and we frequency matched the controls to the cases by sex and age (within ±2 years) at a ratio of 1:1. Hypertension was defined as systolic BP (SBP) ≥140 mmHg and/or diastolic BP (DBP) ≥90 mmHg measured in the supine position. Patients were defined as bedridden if they showed dependency in all sub-items in Katz's ADL (*12*), in addition to being permanently confined to bed (score: 1 or 2) according to the sub-item of "activity" score in the Braden scale (*13*). Patients were defined as non-bedridden if they showed independence in all of Katz's ADL items (*12*). None of the subjects had had any antihypertensive treatment for 2 months prior to enrollment. We excluded 1) subjects admitted to the hospital or at the start of the investigation with clinical diagnoses of Parkinsonism (*14*), Shy-Drager syndrome (*15*), amyloidotic polyneuropathy (*16*), or vitamin B_{12} deficiency (*17*); 2) subjects treated with any drug that may have contributed to or decreased the likelihood of orthostatic hypotension, such as diuretics, α-blockers or β-blockers; 3) subjects considered critically ill (*18*); 4) postoperative patients; and 5) patients admitted for less than 16 weeks.

Procedure for Modified Head-Up Tilt

Basal BP and PR of the elderly subjects were determined by averaging two determinations of supine BP measured with an automatic cuff-oscillometric BO recorder (HEM-705CP; OMRON Co., Ltd., Kyoto, Japan) after the subjects had rested for more than 30 min in the morning before breakfast. The responses of BP and PR were analyzed after a 2-min, 45 deg head-up tilt with the legs horizontal (0 deg) according to the procedure of Gotshall *et al.* (*19*). BP and PR were measured 2 min after the postural change (*20*). Tilt-induced hypotension was defined as a fall in SBP of 20 mmHg or greater and/or DBP of 10 mmHg or greater according to the consensus statement on the definition of orthostatic hypotension (*21*). The changes in BP and PR with the same head-up

Table 2. Comparison of Blood Pressure and Heart Rate at Time of Modified Head-Up Tilt among Normotensive and Hypertensive Inpatients with Non-Bedridden and Bedridden States in a Geriatric Hospital, and between before and after Azelnidipine Treatment

	NB elderly (n=77)		B elderly (n=77)	
	N (n=39)	H (n=38)	N (n=39)	H (n=38)
Before treatment				
Basal supine position				
Systolic BP (mmHg)	122 (106–136)	155 (139–199)†	119 (92–138)$^{\#}$	155 (146–203)$^{\dagger,\#}$
Diastolic BP (mmHg)	70 (54–89)	79 (64–97)†	72 (37–88)$^{\#}$	81 (58–100)$^{\dagger,\#}$
Heart rate (/min)	71 (53–86)	68 (58–86)	75 (58–106)	78 (56–114)
2 min after head-up tilt				
Systolic BP (mmHg)	124 (105–136)	165 (125–188)†	118 (94–162)$^{\#}$	151 (115–199)$^{\dagger,\#}$
Diastolic BP (mmHg)	72 (52–87)	82 (57–105)†	73 (51–93)$^{\#}$	81 (52–132)
Heart rate (/min)	67 (52–85)	66 (52–91)	74 (58–106)	76 (58–99)
Azelnidipine treatment				
Basal supine position				
Systolic BP (mmHg)	—	124 (108–168)$^{\$}$	—	133 (110–170)$^{\$}$
Diastolic BP (mmHg)	—	76 (63–99)$^{\$}$	—	81 (60–91)$^{\$}$
Heart rate (/min)	—	68 (49–86)	—	69 (45–89)$^{\$}$
2 min after head-up tilt				
Systolic BP (mmHg)	—	129 (114–174)$^{\$}$	—	130 (104–162)$^{\$}$
Diastolic BP (mmHg)	—	76 (57–98)$^{\$}$	—	79 (61–90)
Heart rate (/min)	—	69 (51–86)	—	72 (50–93)

Values are expressed as median (range). BP, blood pressure; N, normotensives; H, hypertensives; NB, non-bedridden; B, bedridden; —, no data. $^{\dagger}p<0.008$, *vs.* NB-N group. $^{\#}p<0.008$, *vs.* NB-H group. $^{\$}p<0.05$, *vs.* before treatment with azelnidipine in the same situation in NB-H or B-H groups.

tilt were also determined at 3 months after the start of administration of azelnidipine at a dose of 4 to 16 mg/day.

Data Collection

Operational definitions of each pre-existing potential risk factor for autonomic dysfunction, including stroke (motor deficit and evidence of cerebral hemispheric infarction on computed tomography and/or magnetic resonance imaging) (*22*, *23*), dementia (Mini-Mental State Examination score ≤23) (*17*), chronic ischemic heart disease (previous myocardial infarction or angina pectoris) (*24*), congestive heart failure (left ventricular ejection fraction <40% on echocardiography) (*25*), hypoalbuminemia (serum albumin level <30 g/l) (*26*), and diabetes mellitus (treated with insulin, oral hypoglycemic agent, or fasting blood glucose ≥7 mmol/l) (*26*, *27*), were established prior to data collection. Data were retrieved from medical records before the start of the examination. Personal physicians were involved in the diagnoses of these complications, which were further evaluated by a committee. Objective and routinely collected medical information was applied to augment the diagnostic accuracy. Only chronic conditions were recorded for the cases and respective controls.

Statistical Analyses

Data are expressed as the median and full range. The data were analyzed by Kruskal-Wallis χ^2 and Mann-Whitney U analysis as a multiple comparison using post hoc Bonferroni correction. p values were set at 0.008. Differences in changes of BP and PR by treatment with azelnidipine were assessed by nonparametric Wilcoxon test, and a value of $p<0.05$ was regarded as significant. Data were analyzed on a microcomputer running SPSS version 12 (SPSS, Chicago, USA).

Results

Clinical Characteristics of the Four Groups of Elderly Inpatients

Clinical characteristics of the subjects are shown in Table 1. Numbers of non-bedridden (NB) and bedridden (B) normotensive (N) elderly subjects were 39 (each 12 male and 27 female) and NB and B hypertensive (H) elderly subjects were 38 (each 12 male and 26 female). The median age was not significantly different among the four groups. The prevalence of dementia and chronic phase of stroke in the two bedridden groups (B-N and B-H) was significantly higher than that in

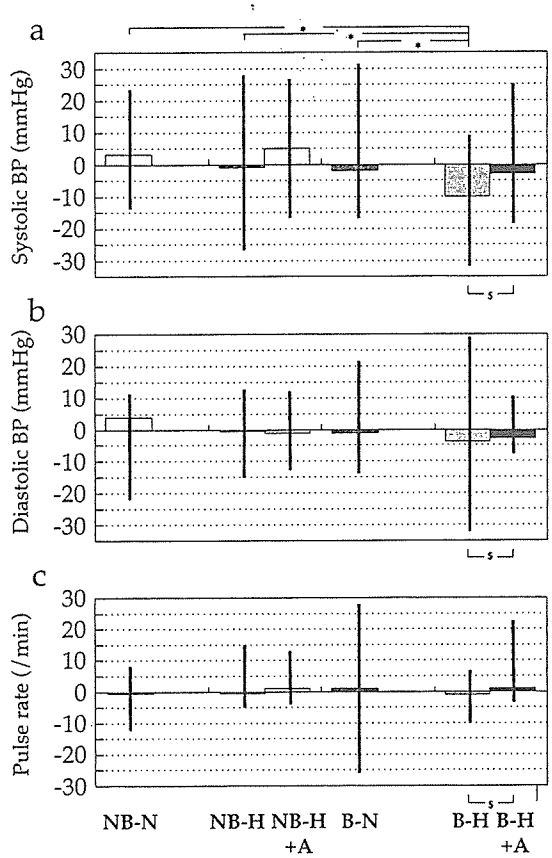

Fig. 1. *Changes in systolic blood pressure (SBP) (a) and diastolic blood pressure (DBP) (b) values and pulse rate (c) at 2 min after head-up tilt from the initial supine position in the four groups. Columns and bars represent the medians and full ranges in each group. NB-N, non-bedridden normotensive group; NB-H, non-bedridden hypertensive group; B-N, bedridden normotensive group; and B-H, bedridden hypertensive group. +A, after treatment with azelnidipine in groups NB-H and B-H.* *$p < 0.008$ vs. *the NB-H group.* $^{\$}p < 0.05$ vs. *before treatment with azelnidipine after the same maneuver in the NB-H or B-H groups.*

the non-bedridden groups (NB-N and NB-H), respectively, and the prevalence of hypoalbuminemia in the B-N group was significantly higher than that in the NB-H group (Table 1).

Changes in BP and PR by Modified Tilt in the Four Groups

Values in SBP, DBP, and PR at the basal supine position and at 2 min after the modified tilt are summarized in Table 2. Both the SBP and DBP values in the two hypertensive groups were significantly higher than those in the two normotensive groups, with the exception that the DBP in the B-H group at 2 min after the tilt was not significantly different from those of the other three groups.

Figure 1 summarizes the changes in SBP and DBP values and PR at 2 min after head-up tilt from the initial supine position in the four groups. Decreases in SBP in the B-H group (median: −10 mmHg; range: −32 to 9 mmHg) were significantly greater than those in either the NB-N group (4 mmHg, −14 to 24 mmHg, $p<0.001$), NB-H group (−1 mmHg, −26 to 28 mmHg, $p=0.001$) or B-N group (−2 mmHg, −17 to 31 mmHg, $p=0.001$), respectively.

On the other hand, the numbers of subjects with tilt-induced hypotension were not significantly different among the NB-N ($n=7$), NB-H ($n=6$), B-N ($n=5$), and B-H ($n=10$) groups by the Kruskal-Wallis test.

Effects of Azelnidipine on BP Change Induced by Head-Up Tilt

Treatment with azelnidipine, a new calcium channel blocker, not only significantly ($p<0.05$) decreased the SBP and DBP levels in the two groups, but also significantly attenuated the higher basal PR in the B-H group to a value comparable to that in the NB-N group (Table 2). Moreover, azelnidipine not only attenuated the tilt-induced prominent decrease in SBP observed before treatment of patients in the B-H group to a level comparable to that in the B-N group, but also significantly enhanced the tilt-induced change in PR to a value comparable to those in the other groups (Fig. 1).

All 10 patients with tilt-induced hypotension in the B-H group were assessed as not having hypotension after the treatment with azelnidipine ($p=0.003$), with significant increments in the tilt-induced changes in SBP (median, −20 mmHg, and range, [−32 to −7 mmHg], before the treatment; to −11 mmHg [−19 to 3 mmHg] after the treatment; $p=0.008$), in DBP (−10 mmHg [−32 to −7 mmHg] to −3 mmHg [−8 to 2 mmHg]; $p=0.005$), and in PR (−1 bpm [−10 to 7 bpm] to 0 bpm [−4 to 23 bpm]; $p=0.024$). Although administration of azelnidipine to the remainder of the 28 patients without tilt-induced hypotension in the B-H group also significantly enhanced the tilt-induced changes in SBP (−4 mmHg [−18 to 9 mmHg] to −2 mmHg [−16 to 25 mmHg]; $p=0.0014$) and in PR (0 bpm [−3 to 7 bpm] to 1 bpm [−1 to 6 bpm]; $p<0.001$), the increment in tilt-induced change in DBP (−1 mmHg [−9 to 5 mmHg] to 1 mmHg [−8 to 10 mmHg]) was not significant ($p=0.210$). Although 4 out of 6 patients with tilt-induced hypotension in the NB-H group were judged not to have hypotension after the treatment with azelnidipine, this difference was not statistically significant ($p=0.157$). Mann-Whitney U analysis and χ^2 analysis did not reveal any significant difference in the mean age or prevalence of the clinical factors, including male gender, dementia, stroke, ischemic heart disease, congestive heart failure, hypoalbuminemia, diabetes mellitus, or bedridden itself, between the 14 hypertensive elderly subjects (4 in the NB-H group and 10 in the B-H group) who showed improvement of the postural hypotension by azelnidipine and the 2 subjects in the NB-H group without the improvement.

Discussion

Our study demonstrated a greater decrease in SBP in the B-H group compared to the other three groups by a quite commonly used nursing maneuver of 45 deg head-up tilt with the legs horizontal. A greater decrease in BP by the head-up tilt is often seen in elderly hypertensive patients (*28*), and is caused by retarded sympathetic nerve activation mainly due to impaired baroreflex sensitivity (*28*), which cannot adjust decreases in cardiac output and arterial pressure due to redistribution of blood from the thoracic area to the deep intra- and inter-muscular vein of the legs by the tilt stress (*29*). The impaired baroreflex activation in these elderly hypertensive patients is represented at least in part by an inadequately lower increment in baroreflex-mediated PR despite a greater decrease in BP compared to that in normotensive elderly subjects (*30*). The elderly B-H patients in this study were characterized by a decrement of the median PR despite a significant decrement of SBP at the time of the tilt, which may be an ultimate feature of autonomic impairment (Fig. 1), suggesting that impaired baroreflex activation played a role in the prominent decrease in SBP in response to the head-up tilt in the B-H patients. On the other hand, increments in both SBP and DBP in response to head-up tilt were observed in NB-N subjects. This value was comparable to that in young normal subjects at the time of head-up tilt in a previous report (*28*). These findings suggest that normotensive healthy subjects, even at a very old age, show a normal autonomic response to the tilt stress, similar to that in young normal subjects.

In the present study, all the patients in the NB-H and B-H groups were treated with azelnidipine, a newly developed dihydropyridine-type calcium channel antagonist with a slowly developing and long-lasting hypotensive effect characterized by little reflex tachycardia (*9*, *10*, *31*). Treatment with azelnidipine not only significantly decreased basal SBP and DBP in patients in both hypertensive groups, but also significantly attenuated the higher basal PR of patients in the B-H group to a level comparable to that of subjects in the NB-N group (Table 2). This observation is partly compatible with a previous report that azelnidipine significantly decreased PR on 24-h ambulatory monitoring (*9*). Despite the significant decreases in BP and basal PR in the B-H patients, azelnidipine not only significantly attenuated the decrements of both SBP and DBP but also reversed the decrement of PR in response to the tilt (Fig. 1). Moreover, all the hypertensive patients with tilt-induced hypotension in the B-H group were judged as not having hypotension after the treatment with azelnidipine. In addition, these patients showed even greater attenuations of the tilt-induced decrements in SBP and DBP compared to those who were originally diagnosed as not having tilt-induced hypotension in the same group, although we used a rather strict criterion for the definition of tilt-induced hypotension—namely, a 20/10 mmHg or greater decline at the time of tilting compared to the spine BP, which is the criterion for orthostatic hypotension (*32*). These observations indicate that the augmenting effect of azelnidipine on the retarded sympathetic nerve activation was more effectively exerted on elderly B-H subjects who showed more severe decreases in BP by the postural change.

Although many antihypertensive agents are known to improve orthostatic hypotension in elderly hypertensive patients (*33*), the associations between dihydropyridine-type calcium blockers and postural hypotension are rather complicated: calcium blockers themselves sometimes induce postural hypotension (*34*), or have no effect on postural hypotension (*35*). Ferodipine (*36*, *37*) and nifedipine (*33*) even attenuate orthostatic hypotension. However, our study is the first to show a high prevalence of postural hypotension in long-term bedridden hypertensive elderly subjects, and the first to show an azelnidipine-induced improvement in postural hypotension among these subjects. The precise mechanism(s) by which azelnidipine improves the head-up-tilt–induced decreases in BP, especially in elderly B-H patients, is unknown. However, unlike in the case of most other long-acting dihydropyridine calcium channel antagonists, the antihypertensive efficacy of azelnidipine is characterized not only by an absence of reactive tachycardia, but also by suppressing effects on sympathetic nervous activity (*9*, *10*, *31*). The additive beneficial effects of the combination of azelnidipine and an angiotensin blocker have recently been described in a hypertensive rat-heart failure model (*38*). On the other hand, the sympathetic nervous activities in astronauts on the 12th and 13th spaceflights during the Neurolab space shuttle mission were actually enhanced compared to the pre-flight levels (*39*). According to these findings as well as the results on the recording of sympathetic nervous activity in subjects exposed to ground-based short- and long-term "stimulations" of microgravity induced by head-down tilt, Mano (*40*) reported that sympathetic neural control was lowered when subjects were exposed to short-term microgravity for several hours, but was enhanced after exposure to long-term microgravity for more than 3 days. In addition, he reported that the orthostatic intolerance based on impaired baroreflex functions in these subjects may have resulted from an exhaustion of the sympathetic nervous system by prolonged exaggerated sympathetic activity after exposure to long-term microgravity stress. He also indicated that the autonomic dysfunctions seen in bedridden subjects may be medicated by the same mechanism (*40*). Based on these observations, the beneficial effect of azelnidipine on the postural hypotension observed especially in elderly B-H patients may be the result of the augmenting effect of azelnidipine on baroreflex sensitivity through a suppression of the exaggerated sympathetic activity due to long-term microgravitational stress. It is also possible that the enhancing effect of azelnidipine on blood flow of the brain (*41*) may have contributed to the improvement of tilt-induced hypotension, since postural hypotension is associated with ischemic change in the brain on magnetic resonance imaging, such as periventricular white matter hyperintensity,

in older elderly subjects (*5*).

In the present study, we did not evaluate hemodynamic or humoral factors, such as echocardiographic measurements and circulating levels of catecholamines and factors associated with the renin-angiotensin-aldosterone system, at the time of tilting. We also did not estimate baroreflex function. These factors should be measured in the future to elucidate the beneficial effects of azelnidipine.

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