

Para-Hisian Pacing for a Pediatric Patient with a Congenitally Corrected Transposition of the Great Arteries (SLL)

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We report a pediatric patient with a congenitally corrected transposition of the great arteries (ccTGA)(SLL) in which permanent para-Hisian pacing (PPHP) could improve dyssynchrony-associated systemic ventricular (SV) dysfunction resulting from permanent morphologic left ventricular pacing for complete atrioventricular block. Since, in patients with ccTGA(SLL), an elongated His-bundle runs medially toward the upper septum to the site of the fibrous continuity between the right-sided mitral valve and pulmonary artery, the His-bundle may easily be captured by a pacing lead, unlike in normal hearts. Thus, PPHP may be an effective therapeutic strategy for the treatment of dyssynchrony-associated SV dysfunction associated with ccTGA (SLL). (PACE 2010; 33:e4–e7)

transposition of the great arteries, para-Hisian pacing, heart failure, cardiac resynchronization therapy, pediatrics, dyssynchrony

Introduction

The cardiac-resynchronization therapy (CRT) utilizing tranvenous biventricular pacing has become proven to produce clinical benefits in adult¹ and pediatric² patients with dyssynchrony-associated systemic ventricular (SV) dysfunction. Here, we report a case of prevention of ventricular desynchronization, so-called cardiac resynchronization, by utilizing tranvenous permanent para-Hisian pacing (TPPHP) for dyssynchrony-associated SV dysfunction resulting from permanent morphologic left ventricular (mLV) pacing for complete atrioventricular (AV) block (CAVB) in a pediatric patient with a congenitally corrected transposition of the great arteries (ccTGA)(SLL).

Case Report

An 8-year-old girl was admitted to our hospital with a chief complaint of general fatigue and impaired daily life activities under appropriate medication with an angiotensin II receptor blocker, β -adrenergic receptor blocker, and diuretics. She had a history of repeated cardiac surg-

eries, consisting of a pulmonary artery banding for ccTGA(SLL), and coexisting anomalies involving an atrial septal defect (ASD) and ventricular septal defect (VSD) at a month after her birth (Fig. 1A), and the implantation of permanent epicardial pacing leads into the right atrium (RA), mLV, and SV, respectively, for CAVB following both an ASD and VSD closure at the age of 1 year (Figs. 1B and 2). The pulse generator was implanted in the intraperitoneum. Although the mLV and RA leads were still functional, the SV lead had already fractured (Fig. 2).

On admission, her blood pressure was 96/58 mmHg, and auscultation revealed a significant regurgitant systolic murmur. The electrocardiogram (ECG) revealed all ventricular pacing with a prolonged QRS duration following the intrinsic P wave (Fig. 1C). Chest X-rays revealed cardiomegaly with a 60% cardio-thoracic ratio. Her initial two-dimensional and Doppler echocardiography (UCG) revealed a severe dyssynchronous SV (morphologic RV) contraction with an SV dilation, reduced SV ejection fraction (SVEF = 19%), and severe left-sided AV (tricuspid, left) valvular regurgitation (Table I). The serum brain natriuretic peptide (BNP) was elevated to 263 pg/dL. Since we presumed the permanent mLV pacing might contribute to her SV dysfunction and heart failure (HF) progression, with left-sided AV valve regurgitation, preventative therapy with ventricular desynchronization was considered. We performed TPPHP for prevention of ventricular desynchronization, following an electrophysiological study, under general anesthesia. The bipolar screw-in leads were advanced via the left subclavian vein

There are no conflicts of interest that are related to the manuscript.

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PARA-HISIAN PACING FOR ccTGA

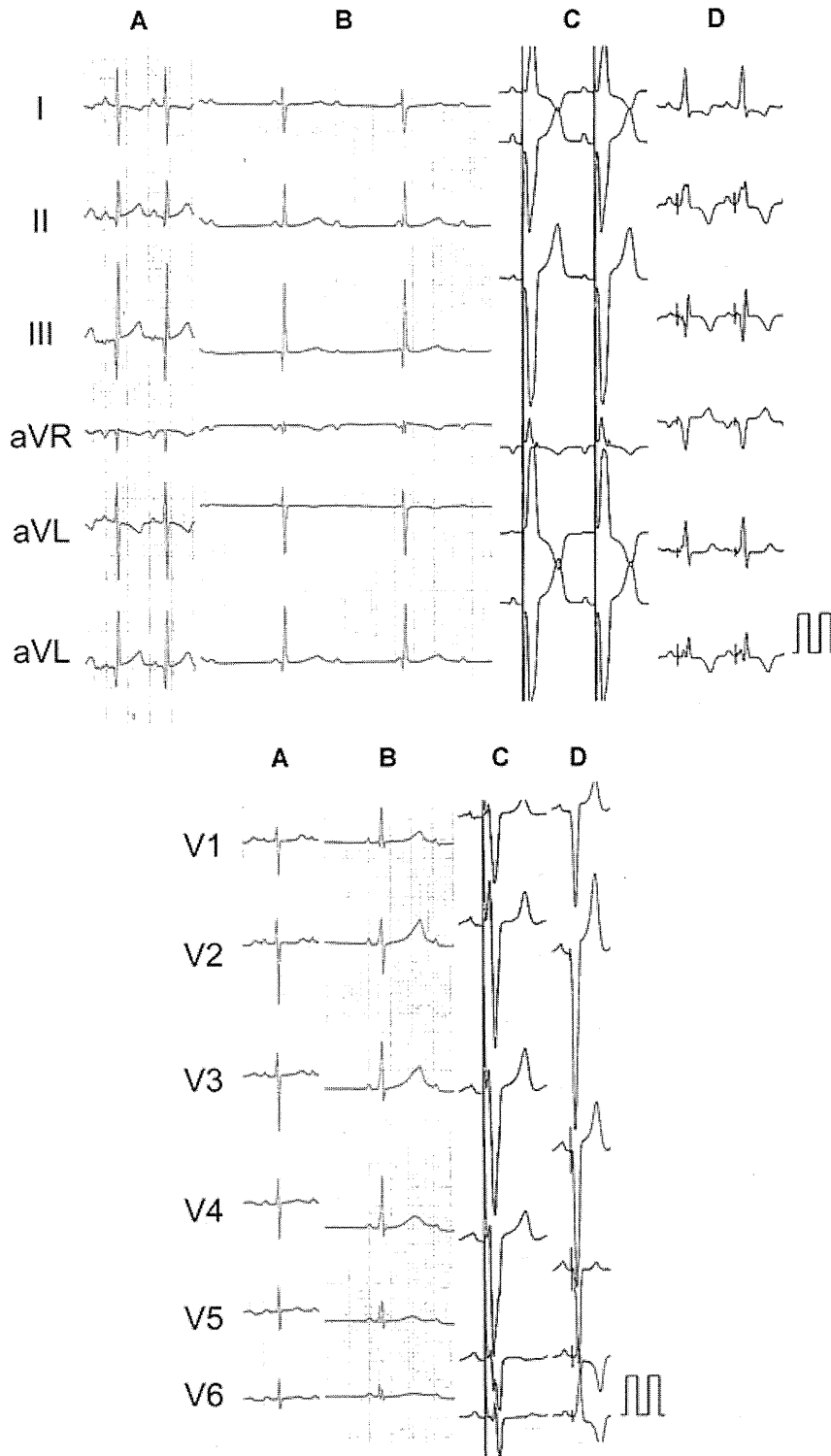


Figure 1. The electrocardiogram during sinus rhythm at a month after her birth (A), during complete atrioventricular block at an age of 1 year (B), during epicardial morphologic left ventricular pacing on admission (QRS duration = 198 ms)(C), and during para-Hisian pacing (QRS duration = 94 ms)(D).

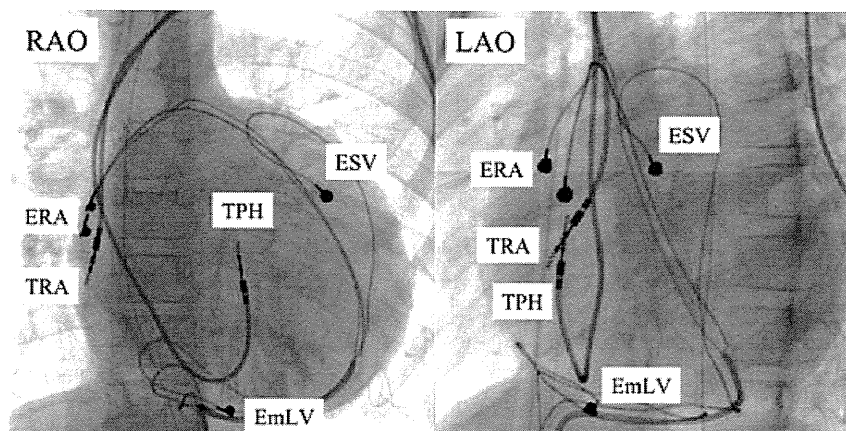


Figure 2. Fluoroscopic images in the right (left panel) and left (right panel) anterior oblique (RAO and LAO) views. The EmLV, ERA, and ESV indicate the epicardial lead(s) in the morphologic left ventricle, right atrium, and systemic ventricle, respectively. The TPH and TRA indicate the transvenous para-Hisian pacing and transvenous right atrial leads, respectively.

and easily positioned, using a steerable stylet into the para-His-bundle in the interventricular septum (Medtronic 5076-58, Medtronic Japan Co., Ltd, Tokyo, Japan) and RA (Medtronic 5076-52), respectively (Fig. 2), with the pulse generator (Guidant INSIGNIA I Plus DR, Guidant Japan KK, Tokyo, Japan) placed in the upper area anterior to the pectoralis major muscle of the left chest, as previously described.³ Further, the pulse generator implanted in the intraperitoneum was turned off thereafter. The pacing threshold of the para-His-bundle lead was 0.7 Volt. The AV delay was optimized at 120 ms by UCG. TPPHP exhibited a concordant QRS axis with a native QRS (Fig. 1A and D), and could dramatically shorten the QRS duration of the ECG (Fig. 1C and D) and decrease the degree of left-sided AV valvular regurgitation as determined by UCG in accordance with an improvement in the dyssynchronous SV contraction (Table I). These conditions met the requirements of a successful criteria for TPPHP as previously defined.⁴ Finally, her daily life activities steadily improved thereafter. Six months after the procedures, her clinical status, cardiomegaly, SVEF, dyssynchronous SV contraction, and serum BNP level all improved (Table I).

Discussion

Recent clinical trials have revealed that permanent right ventricular pacing could promote a dyssynchronous ventricular contraction leading to SV dysfunction and HF progression in adult patients,⁵ and also contribute to adverse histological remodeling and eventual contractile dysfunction in pediatric patients.⁶ A previous clinical study demonstrated that TPPHP may be one therapeutic

strategy for the prevention of ventricular desynchronization in adult patients.⁴

In patients with ccTGA(SLL), the AV node is located well outside Koch's triangle, typically in an anterior and slightly lateral location within the RA, and an elongated His-bundle runs

Table I.

Parameters before and after Para-Hisian Pacing

	Before	After
Echocardiographical evaluation		
Systemic ventricular ejection fraction (%)	19	25
End-diastolic diameter of the systemic ventricle (mm)	64	63
Left-sided atrioventricular valve regurgitation (degree)	4	1
Interventricular delay ⁸ (milliseconds)	137	37
Other parameters		
QRS duration of the electrocardiogram (milliseconds)	198	94
New York Heart Association functional class	III	II
Serum brain natriuretic peptide (pg/dl)	263	151
Cardio-thoracic ratio (%)	60	53

Before, before para-Hisian pacing; After, 6-months after para-Hisian pacing
The interventricular delay was calculated by echocardiography, as previously described.⁸

medially toward the upper septum to the site of the fibrous continuity between the right-sided mitral valve and pulmonary artery in that region.⁷ Thus, the His-bundle may easily be captured by a pacing lead unlike that in normal hearts. At present, the main limiting factor of the transvenous implantation of pacing leads in children may be the existence of the possible risk of a pacing lead distension and/or dislodgment associated with the patients' growth. It was suggested that such risk would be low, since these patients may have a slightly shorter stature; however, TPPHP may be prone to scarring and there are no long-term data on these leads, especially in children.

Thus, we performed TPPHP for prevention of ventricular desynchronization in this present case. It dramatically improved the dyssynchronous SV contraction and clinical status (Table I). In view of these findings, TPPHP may be one of the effective therapeutic strategies for the treatment of dyssynchrony-associated SV dysfunction and HF progression with cCTGA(SLL). Supporting data from large clinical trials, however, are needed before such conclusions can be made.

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Parallel resetting of arterial baroreflex control of renal and cardiac sympathetic nerve activities during upright tilt in rabbits

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Kamiya A, Kawada T, Mizuno M, Shimizu S, Sugimachi M. Parallel resetting of arterial baroreflex control of renal and cardiac sympathetic nerve activities during upright tilt in rabbits. *Am J Physiol Heart Circ Physiol* 298: H1966–H1975, 2010. First published March 26, 2010; doi:10.1152/ajpheart.00340.2009.—Since humans are under ceaseless orthostatic stress, the mechanisms to maintain arterial pressure (AP) against gravitational fluid shift are important. As one mechanism, it was reported that upright tilt reset baroreflex control of renal sympathetic nerve activity (SNA) to a higher SNA in anesthetized rabbits. In the present study, we tested the hypothesis that upright tilt causes a parallel resetting of baroreflex control of renal and cardiac SNAs in anesthetized rabbits. In anesthetized rabbits ($n = 8$, vagotomized and aortic denervated) with 0° supine and 60° upright tilt postures, renal and cardiac SNAs were simultaneously recorded while isolated intracarotid sinus pressure (CSP) was increased stepwise from 40 to 160 mmHg with increments of 20 mmHg. Upright tilt shifted the reverse-sigmoidal curve of the CSP-SNA relationship to higher SNA similarly in renal and cardiac SNAs. Although upright tilt increased the maximal gain, the response range and the minimum value of SNA, the curves were almost superimposable in these SNAs regardless of postures. Scatter plotting of cardiac SNA over renal SNA during the stepwise changes in CSP was close to the line of identity in 0° supine and 60° upright tilt postures. In addition, upright tilt also shifted the reverse-sigmoidal curve of the CSP-heart rate relationship to a higher heart rate, with increases in the maximal gain and the response range. In conclusion, upright posture caused a resetting of arterial baroreflex control of SNA similarly in renal and cardiac SNAs in anesthetized rabbits.

blood pressure; orthostasis; sympathetic nervous system

SINCE HUMANS ARE UNDER CEASELESS orthostatic stress, the mechanisms to maintain arterial pressure (AP) against gravitational fluid shift are greatly important. During standing, a gravitational fluid shift directed toward the lower part of the body (such as the abdominal vascular bed and lower limbs) will cause severe postural hypotension if not counteracted by compensatory mechanisms (15). Arterial baroreflexes have been considered to be the major compensatory mechanism (1, 13, 15), since denervation of baroreceptor afferents causes profound postural hypotension (16). In addition, we (8) recently reported that upright tilt resets baroreflex control of sympathetic nerve activity (SNA) to higher SNA. The resetting doubles SNA, compensates for the reduced pressor responses of cardiovascular organs to SNA during gravitational stress, and contributes to prevent postural hypotension. However, since the study recorded only renal SNA, it remains unknown whether upright tilt resets arterial baroreflex control of SNA innervating cardiovascular organs (i.e., the heart) other than the

kidney. Since cardiac SNA has a critical role in circulation, baroreflex control of cardiac SNA during orthostatic stress is of importance.

Accordingly, in the present study, we tested the hypothesis that upright tilt causes a parallel resetting of arterial baroreflex control of renal and cardiac SNAs in anesthetized rabbits. Since total baroreflex is a closed-loop negative feedback system that senses baroreceptor pressure and controls AP, and since the baroreflex control of SNA (from baroreceptor pressure input to SNA) is a subsystem of the total baroreflex system, it is difficult to isolate the baroreflex control of SNA from the total system in the baroreflex closed-loop condition (16). Therefore, we opened the baroreflex feedback loop by vascularly isolating the carotid sinus region and loaded artificial stepwise intracarotid sinus pressure (CSP) in anesthetized rabbits. By recording renal and cardiac SNAs simultaneously, we investigated static characteristics of baroreflex control of these SNAs (CSP-SNA relationship) in 0° and 60° upright tilt postures.

METHODS

Animals, preparation, and measurements. Japanese White rabbits weighing 2.4–3.3 kg were used. Animals were cared for in strict accordance with the “Guiding Principles for the Care and Use of Animals in the Field of Physiological Science” approved by the Physiological Society of Japan.

Animals ($n = 8$) were initially anesthetized by intravenous injection (2 ml/kg) of a mixture of urethane (250 mg/ml) and α -chloralose (40 mg/ml). Anesthesia was maintained by continuously infusing the anesthetics at a rate of 0.33 ml·kg⁻¹·h⁻¹ using a syringe pump (CFV-3200; Nihon Kohden, Tokyo, Japan). The rabbits were mechanically ventilated with oxygen-enriched room air. Bilateral carotid sinuses were isolated vascularly from the systemic circulation by ligating the internal and external carotid arteries and other small branches originating from the carotid sinus regions. The isolated carotid sinuses were filled with warmed physiological saline, pre-equilibrated with atmospheric air, through catheters inserted via the common carotid arteries. The intracarotid sinus pressure (CSP) was controlled by a servo-controlled piston pump (model ET-126A; Labworks, Costa Mesa, CA). Bilateral vagal and aortic depressor nerves were sectioned in the middle of the neck region to eliminate reflexes from the cardiopulmonary region and the aortic arch. The systemic AP was measured using a high-fidelity pressure transducer (Millar Instruments, Houston, TX) inserted retrograde from the right common carotid artery below the isolated carotid sinus region. Heart rate (HR) was measured with a cardiometer (model N4778; San-ei, Tokyo, Japan).

Body temperature was maintained at around 38°C with a heating pad. The left renal sympathetic nerve was exposed retroperitoneally, and the left cardiac sympathetic nerve was exposed through a middle thoracotomy. A pair of stainless steel wire electrodes (Bioflex wire AS633; Cooner Wire) was hooked onto each of these nerves to record renal and cardiac SNAs. The nerve fibers peripheral to electrodes were ligated securely and crushed to eliminate afferent signals. The nerve

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and electrodes were covered with a mixture of silicone gel (silicon low viscosity, Kwik-Sil; World Precision Instruments, Sarasota, FL) to insulate and immobilize the electrodes. The preamplified SNA signals were band-pass filtered at 150–1,000 Hz. These nerve signals were full-wave rectified and low-pass filtered with a cutoff frequency of 30 Hz to quantify the nerve activity. After the experiment, an intravenous infusion of hexamethonium bromide (6 mg/kg) abolished the SNA signals, indicating that the signals recorded were postganglionic SNA.

Protocols. After the preparation, the animal was maintained in a 0° supine posture on a tilt bed. To stabilize the posture, the head was fixed full-frontal to the bed by strings, and the body and legs were rigged up in a clothes-like bag. Bilateral CSP was artificially controlled independently of systemic AP. First, actual operating pressure and SNAs under baroreflex closed-loop conditions in the 0° supine posture were obtained. The animal was kept in the 0° supine posture for 10 min while CSP was matched with systemic AP via the servo-controlled piston pump.

Second, the static characteristics of the sympathetic baroreflex system were estimated in the 0° position under baroreflex open-loop conditions. The animal was kept in the 0° supine posture while CSP was decreased to 40 mmHg and then increased stepwise from 40 to 160 mmHg in increments of 20 mmHg. Each CSP step was maintained for 60 s.

Third, actual operating pressure and SNAs under baroreflex closed-loop conditions in the 60° upright tilt position were obtained. The animal was kept supine for 10 min and then tilted upright to 60° within 10 s by inclining the tilt bed to 60° and dropping the lower regions of the rabbit with the fulcrum set at the level of the carotid sinus. The 60° upright posture was maintained for 10 min. CSP was matched with systemic AP via the servo-controlled piston pump.

Since the clothes-like bag stabilized the posture of the animals, there was no additional mechanical movement that reduced the quality of measurements. The position of the head remained almost fixed during the tilt to minimize vestibular stimulation. Last, the static characteristics of the sympathetic baroreflex system were estimated during the 60° upright tilt posture. CSP was increased stepwise from 40 to 160 mmHg similarly to the experiment in the 0° position. These SNAs and AP were recorded at a 200-Hz sampling rate using a 12-bit analog-to-digital converter and stored on the hard disk of a dedicated laboratory computer system for later analysis.

Data analysis. The SNA signals were normalized by the following steps. First, for each type of SNA, 0 arbitrary unit (a.u.) was assigned to the postmortem noise level. Second, 100 a.u. were assigned to the average of actual operating SNA values during baseline period in 0° positions. Last, the other SNA signals were then normalized to these values in each experiment.

These SNA and HR values were averaged for the last 10 s of each CSP level. The static relationships between CSP and SNA and between CSP and HR were parameterized by two widely used traditional models (nonlinear reverse-sigmoidal curve, linear regression line), although both models have limited abilities to reproduce the actual data. In the former case, the data were parameterized by a four-parameter logistic equation model as follows:

$$Y = P_4 + \frac{P_1}{1 + \exp[P_2(CSP - P_3)]} \quad (1)$$

where Y is SNA or HR, P_1 is the response range of Y (i.e., the difference between the maximum and minimum values of Y), P_2 is the coefficient of gain, P_3 is the midpoint CSP of the logistic function, and P_4 is the minimum value of Y . We calculated the instantaneous gain

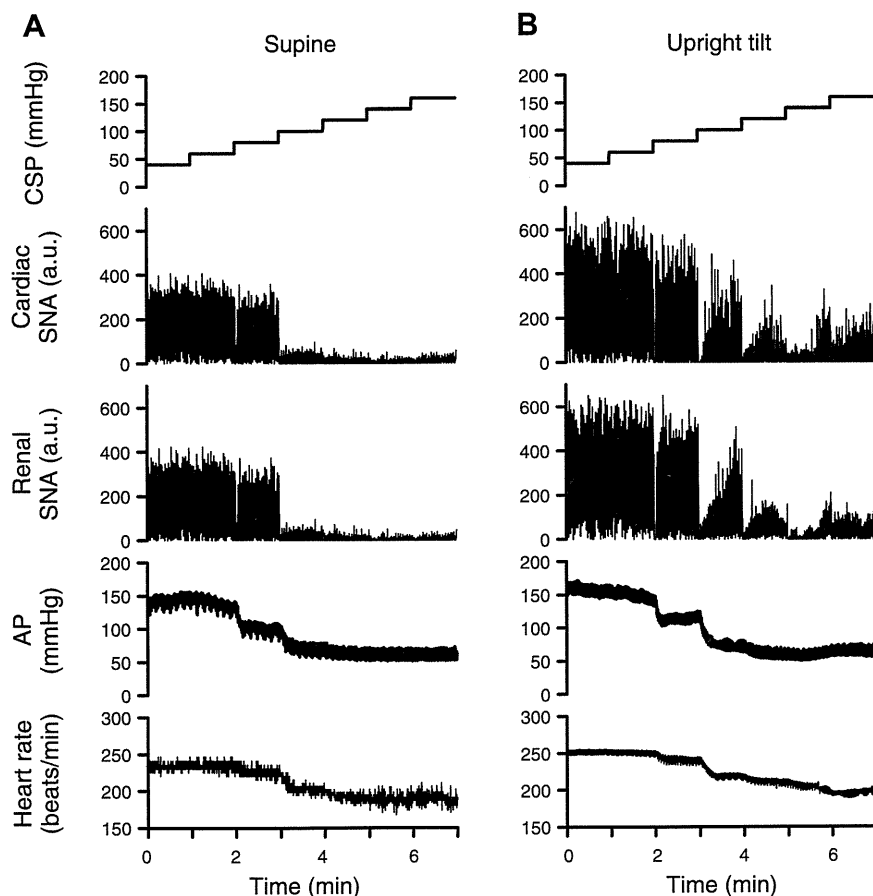


Fig. 1. Representative time series of renal and cardiac sympathetic nerve activities (SNAs) and arterial pressure (AP) in response to a stepwise increase in intracarotid sinus pressure (CSP) in 0° supine (A) and 60° upright tilt postures (B) obtained from 1 animal. Each CSP step was maintained for 1 min. All data were sampled at 10 Hz. In both SNAs, increasing CSP decreased SNAs in both postures, but upright tilt increased SNAs at all CSP levels. a.u., Arbitrary unit.

from the first derivative of the logistic function and the maximum gain (G_{max}) from $-P_1P_2/4$ at $CSP = P_3$.

Statistic analysis. All data are means \pm SD. Effects of the upright tilt on baroreflex parameters were evaluated by repeated-measures analysis of variance. When the main effect was found to be significant, post hoc multiple comparisons were made using Scheffé's *F*-test to compare baroreflex controls between renal and cardiac SNAs (3). Differences were considered significant when $P < 0.05$.

RESULTS

Baroreflex control of renal and cardiac SNAs. Figure 1 shows the representative time series data obtained from one subject. The renal and cardiac SNAs similarly decreased in response to stepwise increase in CSP in the 0° supine posture (Fig. 1A). The 60° upright tilt increased these SNAs at each CSP level (Fig. 1B) compared with the supine posture.

Figure 2 shows the relationship between CSP and SNA in the same data as in Fig. 1A. In Fig. 2, these SNAs were averaged for the last 10 s of each CSP level to investigate the steady-state, not transient, response to a stepwise change in CSP. The 60° upright tilt increased renal (Fig. 2A) and cardiac SNAs (Fig. 2C) at all CSP levels compared with the supine posture. The renal SNA approximately matched the cardiac SNA at all CSP levels in the supine posture (Fig. 2B) and also in the upright tilt posture (Fig. 2D).

When the static relationship between CSP and each SNA was fitted to a nonlinear reverse-sigmoidal curve (Fig. 3), the r^2 value was ~ 0.95 . The 60° upright tilt shifted the CSP-renal SNA curve upward to a higher SNA (Fig. 3A). Similarly, the upright tilt shifted the CSP-cardiac SNA curve (Fig. 3C) in the same manner as renal SNA. The CSP-renal SNA curve was almost superimposed on the CSP-cardiac SNA curve in the 0° supine (Fig. 3B) and upright tilt postures (Fig. 3D). However, the model was limited in reproducing the data, since the measured SNAs did not saturate at the CSP levels of 40–60 mmHg.

When the static relationship between CSP and each SNA was fitted to a linear regression line (Fig. 4), the r^2 value was 0.82–88, lower than when the nonlinear reverse-sigmoidal curve was used. The 60° upright tilt shifted the CSP-renal SNA (Fig. 4A) and the CSP-cardiac SNA lines (Fig. 4C) upward to the higher SNA levels. The CSP-renal SNA line was almost superimposed on the CSP-cardiac SNA line in the 0° supine (Fig. 4B) and upright tilt postures (Fig. 4D).

Averaged data from all animals showed that the 60° upright tilt increased renal (Fig. 5A) and cardiac SNAs (Fig. 5B) at all CSP levels compared with the 0° supine posture. The renal SNA almost matched the cardiac SNA at all CSP levels in the supine (Fig. 5D) and also in the upright tilt posture (Fig. 5E),

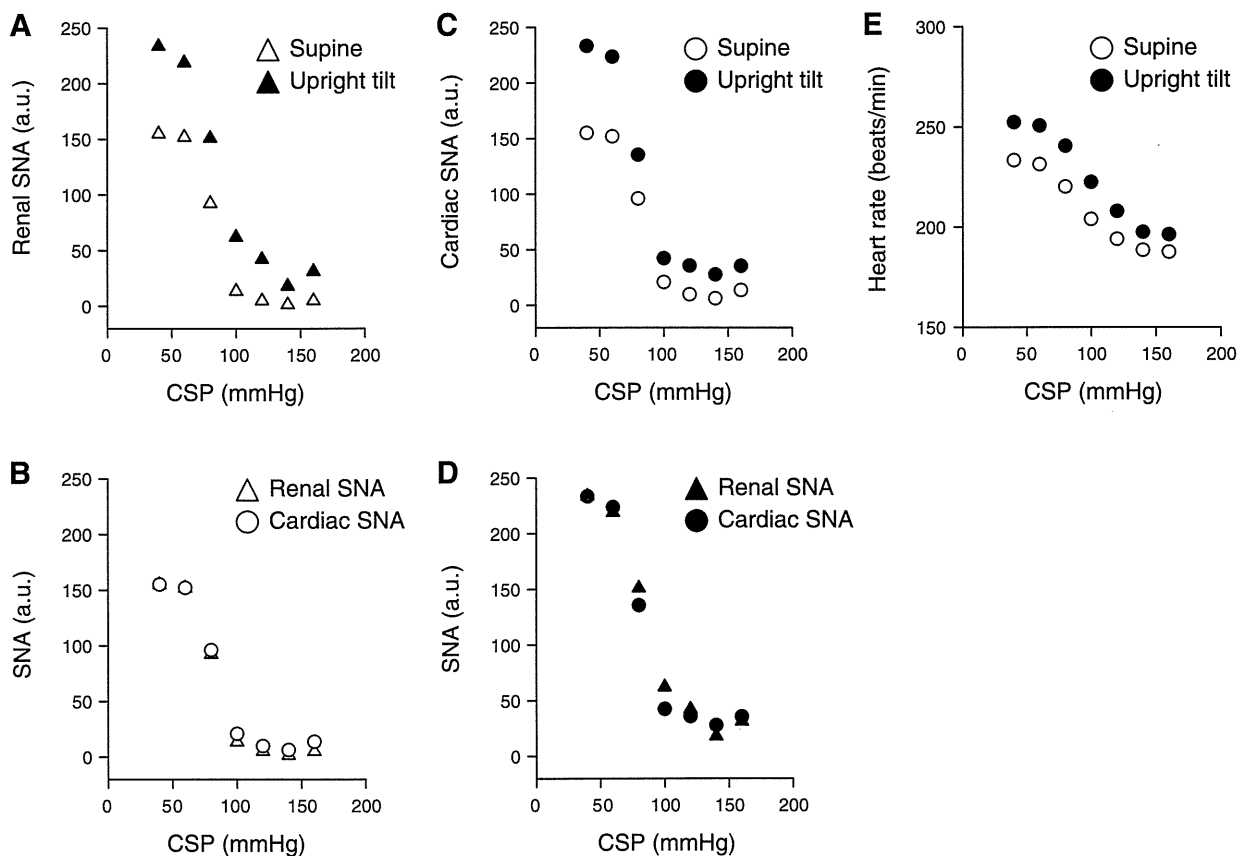


Fig. 2. Example of arterial baroreflex control of renal (A) and cardiac SNAs (C) and heart rate (HR; E). Data were obtained from the same animal studied in Fig. 1 and averaged for the last 10 s of each CSP level. Open and filled symbols show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex control of SNA to a higher SNA similarly in the CSP-renal SNA (A) and CSP-cardiac SNA relationships (C). Data in B and D represent the superimposing of baroreflex control of SNA between renal and cardiac SNAs in both the supine and upright tilt postures, respectively. The upright tilt also shifted the baroreflex control of HR upward (E).

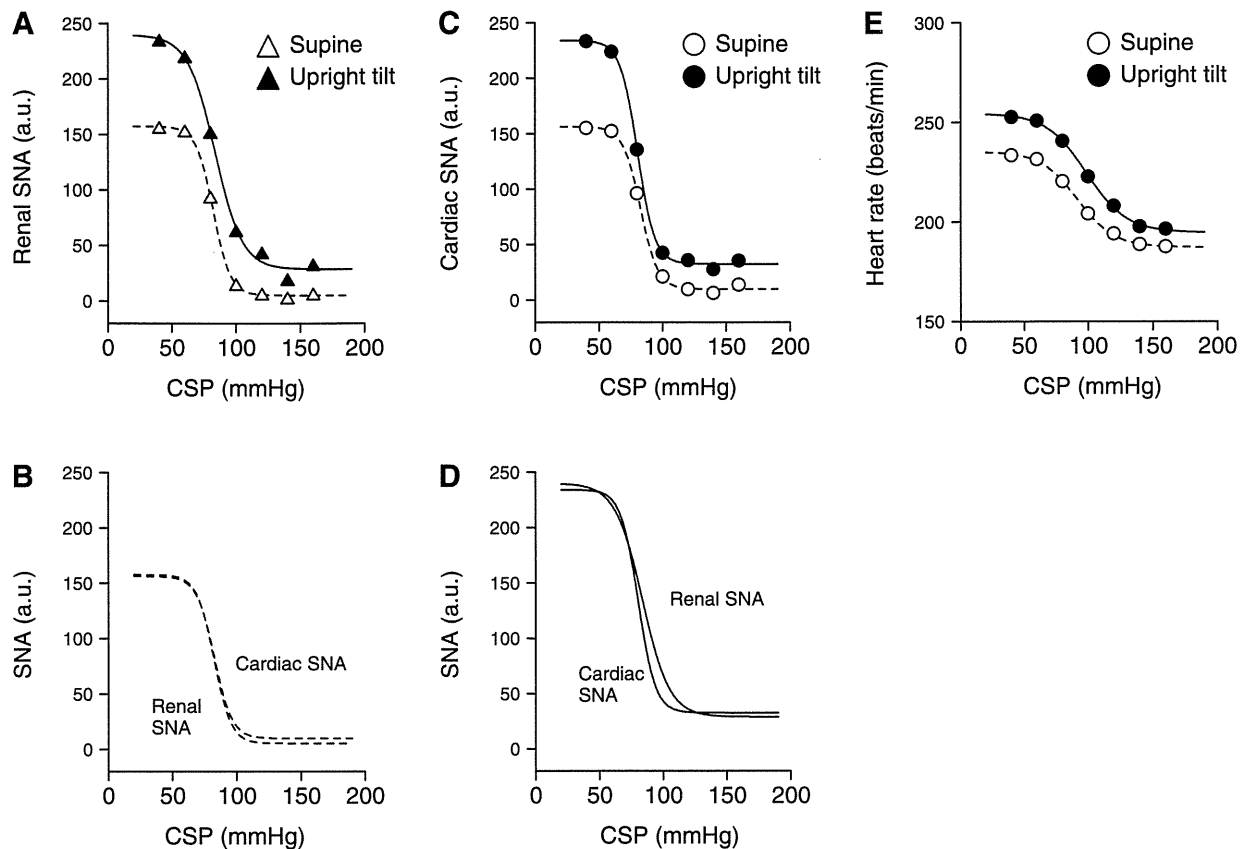


Fig. 3. Example of a model of the data shown in Fig. 2 using reverse-sigmoid 4-parameter logistic functions. Dotted and solid curves show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex curves to a higher SNA similarly in renal (A) and cardiac SNAs (C). The curves were superimposed between these SNAs in the supine (B) and upright tilt postures (D). The upright tilt also shifted the baroreflex curve of HR upward (E).

indicating that 60° upright tilt shifted the CSP-SNA relationship upward by similar magnitudes in renal and cardiac SNAs.

When the static relationship between CSP and each SNA was fitted to a nonlinear reverse-sigmoidal curve (Fig. 6), the upright tilt shifted the CSP-SNA curve to higher SNA similarly in renal (Fig. 6A) and cardiac SNAs (Fig. 6C). The CSP-SNA relationship was almost superimposed between these SNAs in both the supine (Fig. 6B) and upright tilt postures (Fig. 4D). In both renal and cardiac SNAs, P_1 (the range of SNA response to CSP), P_4 (the minimum value of SNA), and the maximal gain (at the midpoint of the logistic function) were larger at upright tilt than supine posture (Table 1), whereas P_2 (the coefficient of gain) and P_3 (the midpoint CSP of the logistic function) were not different between postures (Table 1). In both postures, these parameters of P_{1-4} and maximal gain were similar in renal and cardiac SNAs (Table 1).

When the static relationship between CSP and each SNA was fitted to a linear regression line (Fig. 7), the upright tilt shifted the CSP-SNA line to higher SNA similarly in renal (Fig. 7A) and cardiac SNAs (Fig. 7C). It increased the slope of regression from -1.4 ± 0.3 to -1.8 ± 0.3 a.u./mmHg in renal SNA and from -1.4 ± 0.3 to -1.8 ± 0.4 a.u./mmHg in cardiac SNA. The CSP-SNA lines were almost superimposed on their SNAs in the 0° supine (Fig. 7B) and also the upright tilt posture (Fig. 7D). In both SNAs of all animals, the r^2 value was always lower (0.80–0.89) than when a nonlinear reverse-sigmoidal curve was used (0.92–0.97).

In addition, in both 0° supine and 60° upright tilt postures, scatter plotting of cardiac SNA over renal SNA was approximately close to the line of identity for each subject (Fig. 8A) and the pooled data from all subjects (Fig. 8B), indicating that these SNAs changed in parallel in response to stepwise increase in CSP regardless of posture. The upright tilt did not change operating AP (steady-state AP: 102 ± 4 mmHg in supine posture, 102 ± 5 mmHg in upright tilt posture). The upright tilt increased operating renal (100 a.u. in supine posture, 148 ± 19 a.u. in upright posture) and cardiac SNAs (100 a.u. in supine posture, 155 ± 21 a.u. in upright posture) by similar magnitudes.

Figure 9 showed the discharge characteristics of the renal and cardiac SNAs obtained from the same animal studied in Fig. 1. These SNAs were similar to some extent regardless of baroreflex condition and posture. In the supine posture (Fig. 9A), first, these SNAs were weakly pulse synchronous and had slower fluctuations with a time cycle of ~ 1.7 s in the baroreflex closed-loop condition, where CSP was artificially matched with systemic AP. The CSP and AP also had fluctuations with the same time cycle. Second, in the baroreflex open-loop condition, where CSP was fixed at 40 mmHg (the CSP level was chosen because it maximized these SNAs) without pulse, these SNAs had neither a pulse rhythmicity nor the slower fluctuation observed in the closed-loop condition. These discharge characteristics of SNAs were also observed in the 60° upright posture (Fig. 9B), although the amplitude of SNAs were larger at upright tilt than supine posture.

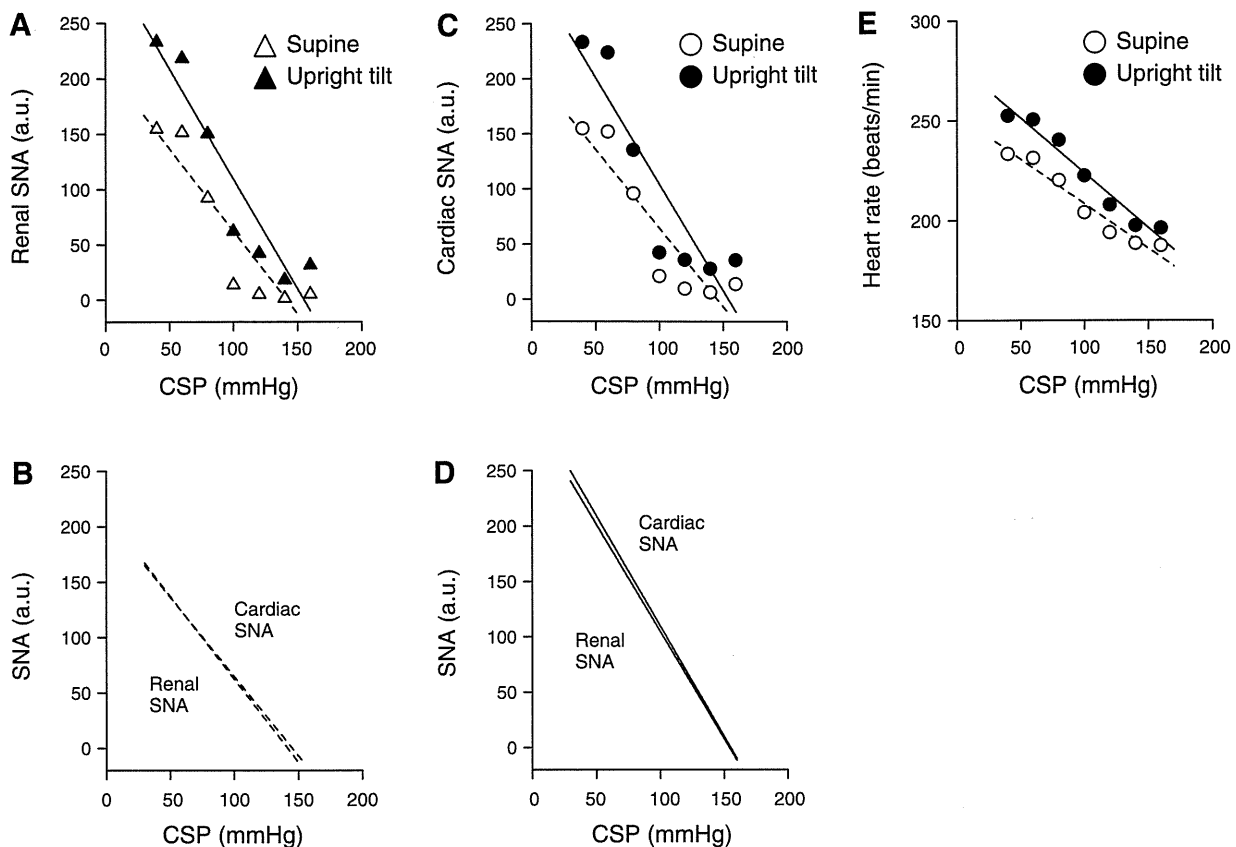


Fig. 4. Example of a model of the data shown in Fig. 2 using a simple regression line. Dotted and solid lines show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex lines to a higher SNA similarly in renal (A) and cardiac SNAs (C). The lines were superimposed between these SNAs in the supine (B) and upright tilt postures (D). The upright tilt also shifted the baroreflex line of HR upward (E).

Baroreflex control of HR. In the representative time-series data, HR decreased in response to a stepwise increase in CSP in the 0° supine posture (Fig. 1A) and during 60° upright tilt (Fig. 1B). The upright tilt shifted the CSP-HR relationship upward to a higher HR (Fig. 2E), although HR was averaged for the last 10 s of each CSP level to investigate the steady-state, not transient, response to stepwise change in CSP.

Averaged data from all animals showed that the upright tilt shifted the CSP-HR relationship upward to a higher HR (Fig. 5E). When the static relationship between CSP and HR was fitted to a nonlinear reverse-sigmoidal curve (Fig. 6E), the P_1 (the range of HR response to CSP) and the maximal gain (at the midpoint of the logistic function) were larger at upright tilt than in the supine posture (Table 2), whereas P_2 (the coefficient of gain), P_3 (the midpoint CSP of the logistic function), and P_4 (the minimum value of HR) were not different between postures (Table 2). When the static relationship between CSP and HR was fitted to a linear regression line (Fig. 8E), the upright tilt increased the slope of regression from 0.46 ± 0.3 to 0.60 ± 0.3 beats \cdot min $^{-1}$ \cdot mmHg $^{-1}$. The upright tilt increased operating HR (steady-state HR; 204 ± 11 beats/min in supine posture, 220 ± 12 beats/min in upright tilt posture).

DISCUSSION

Arterial baroreflex control of SNA is considered to have an important role to maintain AP under orthostatic stress against gravitational fluid shift directed toward the lower part of the

body (15). In addition, we (8) recently reported that upright tilt resets arterial baroreflex control of renal SNA to increase orthostatic sympathetic activation. However, it remains unknown whether upright tilt resets arterial baroreflex control of SNA innervating to cardiovascular organs (i.e., the heart) other than the kidney. One major new finding in this study is that 60° upright tilt resets arterial baroreflex control of SNA to higher SNA similarly in renal and cardiac SNAs. This supports our hypothesis that upright tilt causes a parallel resetting of arterial baroreflex control of renal and cardiac SNAs in anesthetized rabbits.

Some regional differences between renal and cardiac SNAs certainly have been reported under some physiological conditions. First, for example, the dynamic high-pass characteristics in baroreflex control of SNA were greater in cardiac SNA than renal SNA (6, 10). Second, activating left atrial receptors increased cardiac SNA but decreased renal SNA (9). Last, hypoxia reset the AP-SNA relationship to higher AP and SNA in renal SNA but to lower AP and SNA in cardiac SNA (4). These lines of evidence indicate that renal and cardiac SNAs respond differently to specific physiological stimulation and stress (14).

However, our results indicate that upright posture induces a parallel resetting in arterial baroreflex control of renal and cardiac SNAs in the static characteristics. In agreement with previous studies (6, 7), the CSP-renal SNA reverse-sigmoidal curve was superimposable to the CSP-cardiac SNA curve in

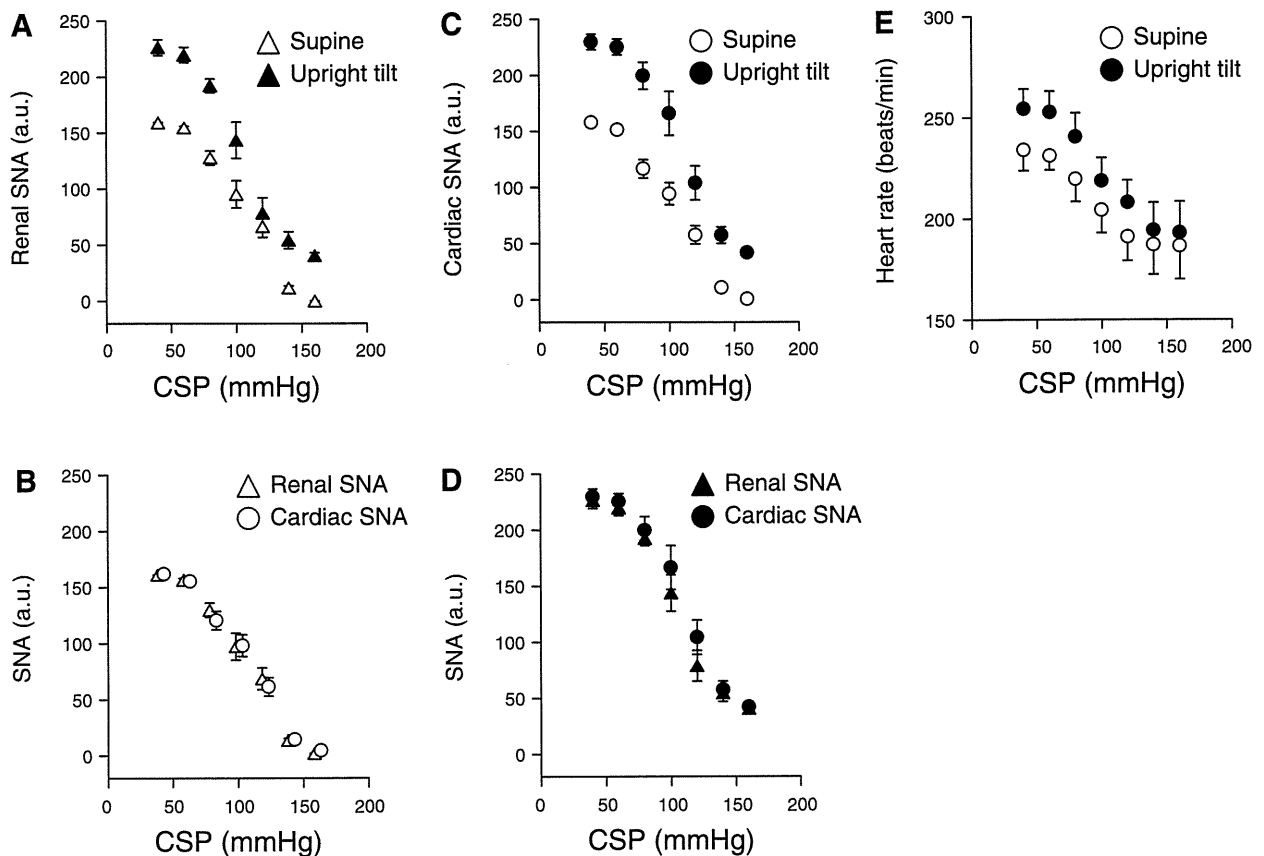


Fig. 5. Averaged data of arterial baroreflex control of renal (A) and cardiac SNAs (C) and HR (E) from all animals ($n = 8$). Open and filled symbols show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex control of SNA to a higher SNA similarly in the CSP-renal SNA (A) and CSP-cardiac SNA relationships (C). B and D represent the superimposing of baroreflex control of SNA between renal and cardiac SNAs in both the supine and upright tilt postures, respectively. The upright tilt also shifted the baroreflex control of HR upward (E). Data are means \pm SD.

the supine posture. This indicates that static nonlinear characteristics in arterial baroreflex control of renal SNA matched those of cardiac SNA in the posture. In addition, since upright tilt posture shifted the CSP-SNA curves upward similarly in renal and cardiac SNAs, the static nonlinear characteristics in arterial baroreflex control of renal SNA also matched those of cardiac SNA in upright tilt posture. These results were consistent with the close correlation between renal and cardiac SNAs during forced CSP changes with supine and upright tilt postures. They might also be consistent with a numerical simulation study indicating parallel responses of renal and cardiac SNAs to physiological pressure perturbations (AP change) (6).

Our results indicate that upright posture resets arterial baroreflex control of HR to a higher HR. This is consistent with the results of baroreflex resetting for cardiac SNA under upright tilt, because the P_1 (the response range) and the maximal gain (at the midpoint of the logistic function) were larger in both CSP-HR and CSP-cardiac SNA relationships. The parallelism suggests that cardiac sympathetic efferent was a dominant determinant for HR in the present experimental condition with cutting of vagal nerves. Our results could be consistent with the increase in the baroreflex gain for HR assessed by a neck pressure/suction device in humans (11).

Limitations. The present study has several limitations. First, we excluded the efferent effect of vagally mediated arterial

baroreflex and an anesthetic agent that could affect baroreflex control of SNA. Second, the vascular isolation of carotid sinus might decrease brain blood flow under, in particular, upright tilt position. Third, we eliminated cardiopulmonary baroreflex by cutting bilateral vagal nerves. Earlier human studies have indicated that nonhypotensive hypovolemic perturbations do not change AP but reduce central venous, right heart, and pulmonary pressures and cause vasoconstriction. These observations have been interpreted as reflexes triggered by cardiopulmonary baroreceptors (5, 12). However, Taylor et al. (17) showed that small reductions of effective blood volume reduce aortic baroreceptive areas and trigger hemodynamic adjustments that are so efficient that alterations in AP escape detection by conventional means. In addition, Fu et al. (2) reported that arterial baroreceptors are consistently unloaded during low levels (i.e., -10 and -15 mmHg) of lower body negative pressure in humans. Accordingly, further studies are needed to understand the relative importance and mutual cooperation of arterial and cardiopulmonary baroreflexes in AP control during orthostatic stress. Fourth, we investigated arterial baroreflex during upright posture in rabbits, which are quadrupeds. However, denervation of both carotid and aortic arterial baroreflexes caused postural hypotension of ~ 50 mmHg during 60° upright tilt in quadrupeds [rabbits and rats (16)]. This suggests that even in quadrupeds, arterial baroreflex has a very important role in maintenance of AP under orthostatic stress.

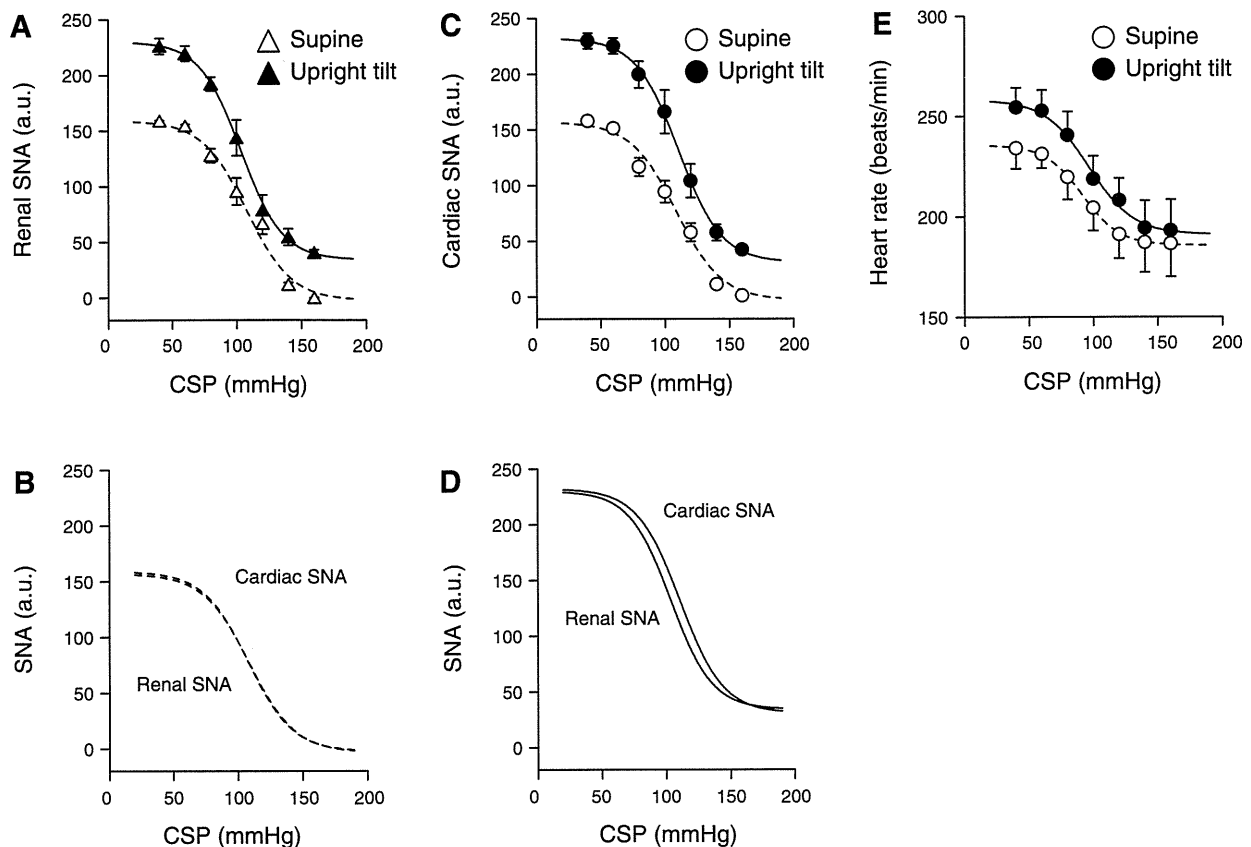


Fig. 6. A model of the averaged data shown in Fig. 5 using reverse-sigmoid 4-parameter logistic functions. Dotted and solid curves show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex curves to a higher SNA similarly in renal (A) and cardiac SNAs (C). The curves were superimposed between these SNAs in the supine (B) and upright tilt postures (D). The upright tilt also shifted the baroreflex curve of HR upward (E).

Last, although we used two widely used traditional models to analyze the relationship between CSP and SNA, both have limited abilities to reproduce actual data. The nonlinear reverse-sigmoidal curve parameterized by a four-parameter logistic equation model provided high r^2 values (0.92–0.97) regardless of SNA type and posture. However, we failed to observe a saturation of SNA at the lowest CSP level in some cases (40 mmHg; Fig. 3, A and B, in upright tilt position). Lots

Table 1. Effect of upright tilt on parameters of baroreflex control of renal and cardiac SNAs

	Supine	Upright tilt
Renal SNA		
P_1 , a.u.	161 ± 2	$196 \pm 5^*$
P_2 , a.u./mmHg	0.08 ± 0.01	0.08 ± 0.02
P_3 , mmHg	105 ± 6	104 ± 6
P_4 , a.u.	2 ± 1	$34 \pm 6^*$
G_{max} , a.u./mmHg	-1.5 ± 0.4	$-1.9 \pm 0.4^*$
Cardiac SNA		
P_1 , a.u.	160 ± 2	$201 \pm 5^*$
P_2 , a.u./mmHg	0.08 ± 0.01	0.08 ± 0.02
P_3 , mmHg	109 ± 6	111 ± 6
P_4 , a.u.	2 ± 1	$31 \pm 6^*$
G_{max} , a.u./mmHg	-1.4 ± 0.4	$-1.9 \pm 0.4^*$

Values are means \pm SD ($n = 8$) for the parameters of baroreflex control of renal and cardiac sympathetic nerve activities (SNAs). See Eq. 1 in METHODS for definitions of the 4 parameters of the logistic function. * $P < 0.05$, supine vs. upright tilt.

of earlier studies have applied the model to AP and SNA (or HR) data under pharmacological perturbation (i.e., nitroprusside, phenylephrine) (1, 14), although it is difficult to observe clear saturation and/or threshold in the data. In contrary, the simple linear regression line model provided lower r^2 values (0.80–0.89). The plotted data did not appear to lie on a simple line in individuals (Fig. 4). Accordingly, we cannot conclude whether the relation between CSP and SNA is sigmoid or not. This problem is not the purpose of this study. Importantly, without modeling, our data (Fig. 2 and 5) indicate the parallel resetting of arterial baroreflex control of renal and cardiac SNAs.

In conclusion, upright posture causes a resetting in arterial baroreflex control of SNA in parallel in renal and cardiac SNAs in anesthetized rabbits.

GRANTS

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

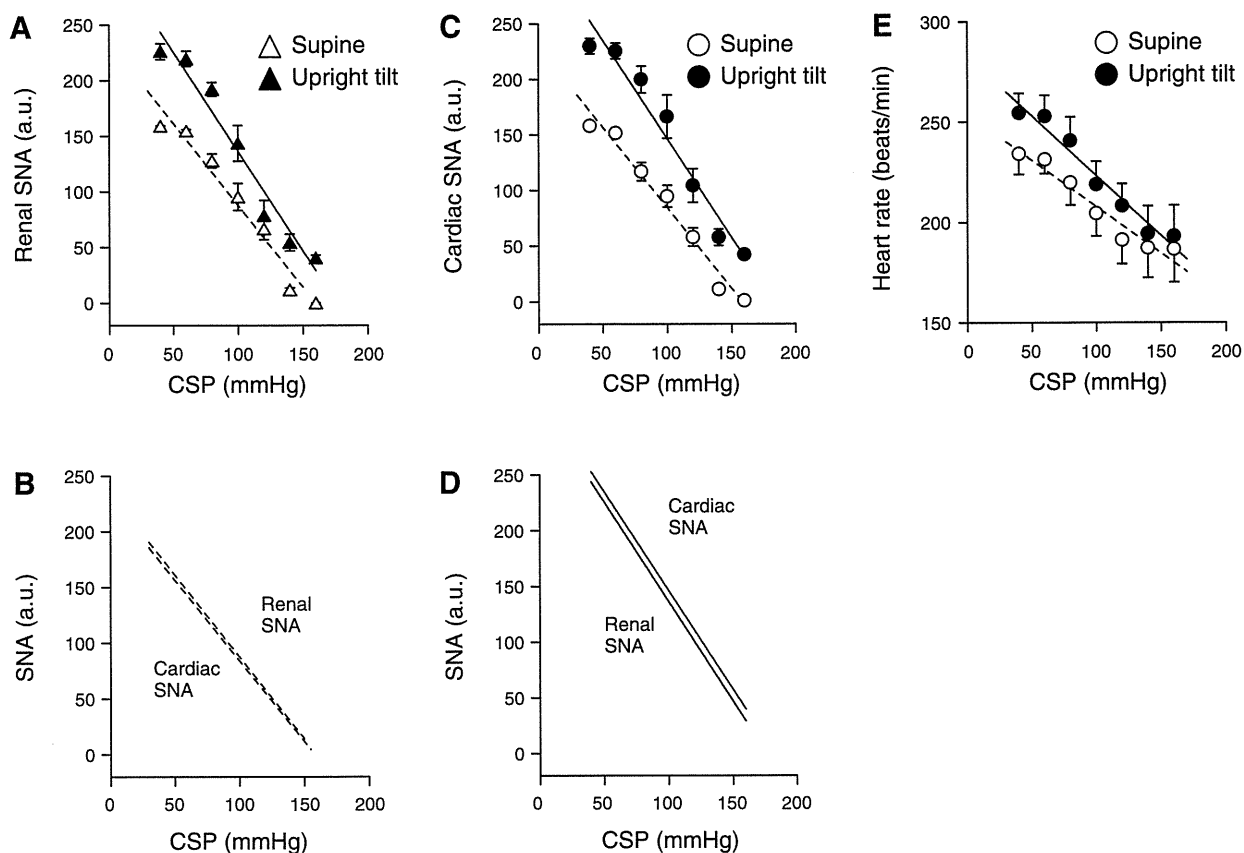


Fig. 7. A model of the averaged data shown in Fig. 5 using a simple regression line. Dotted and solid lines show the data in the supine and 60° upright tilt postures, respectively. The upright tilt shifted the baroreflex lines to a higher SNA similarly in renal (A) and cardiac SNAs (C). The lines were superimposed between these SNAs in the supine (B) and upright tilt postures (D). The upright tilt also shifted the baroreflex line of HR upward (E).

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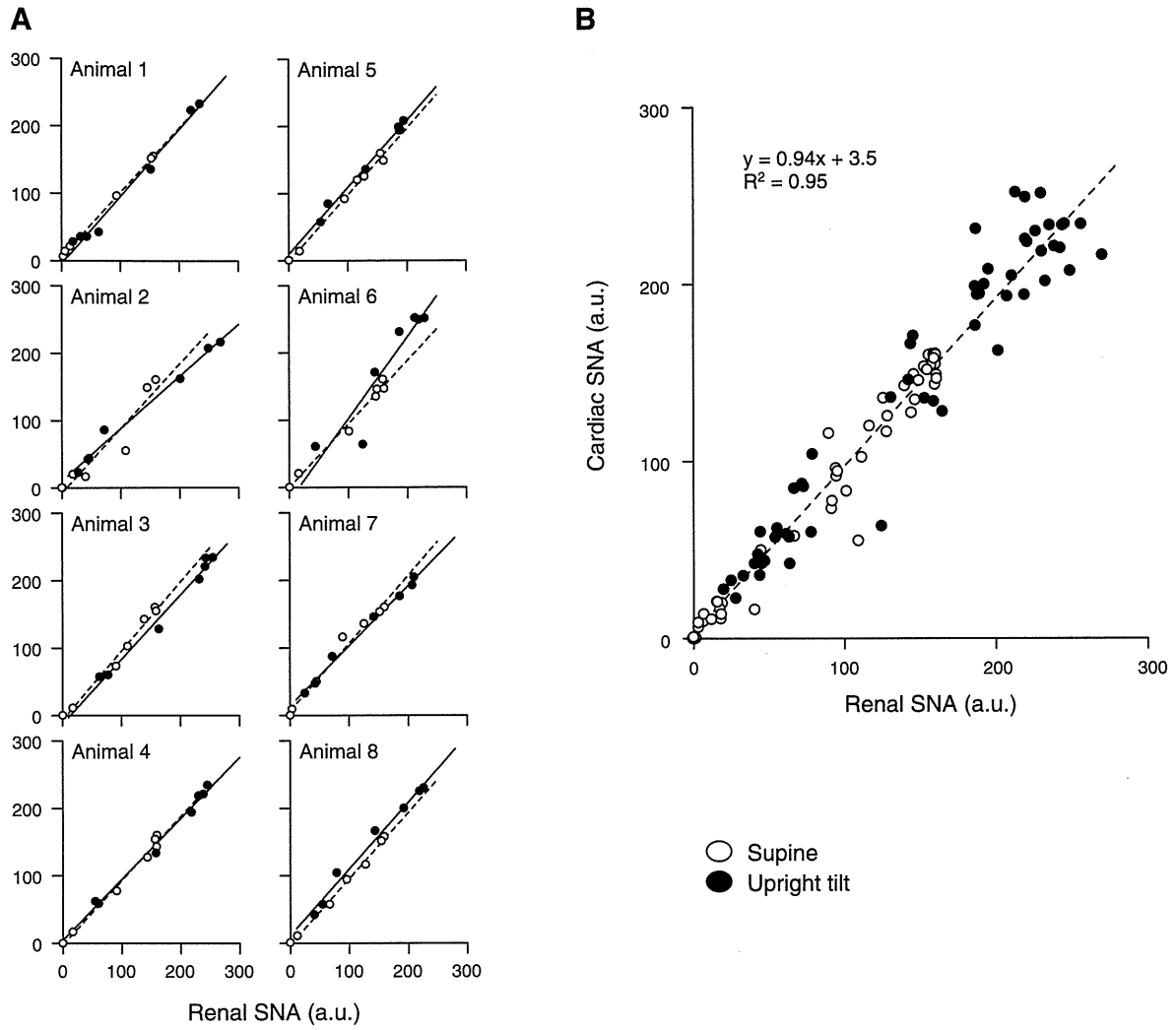


Fig. 8. Scatter plots and regression lines drawn between renal and cardiac SNAs in supine (dotted lines) and upright tilt postures (solid lines) during stepwise changes in CSP for each subject (A) and the pooled data from all 8 subjects (B).

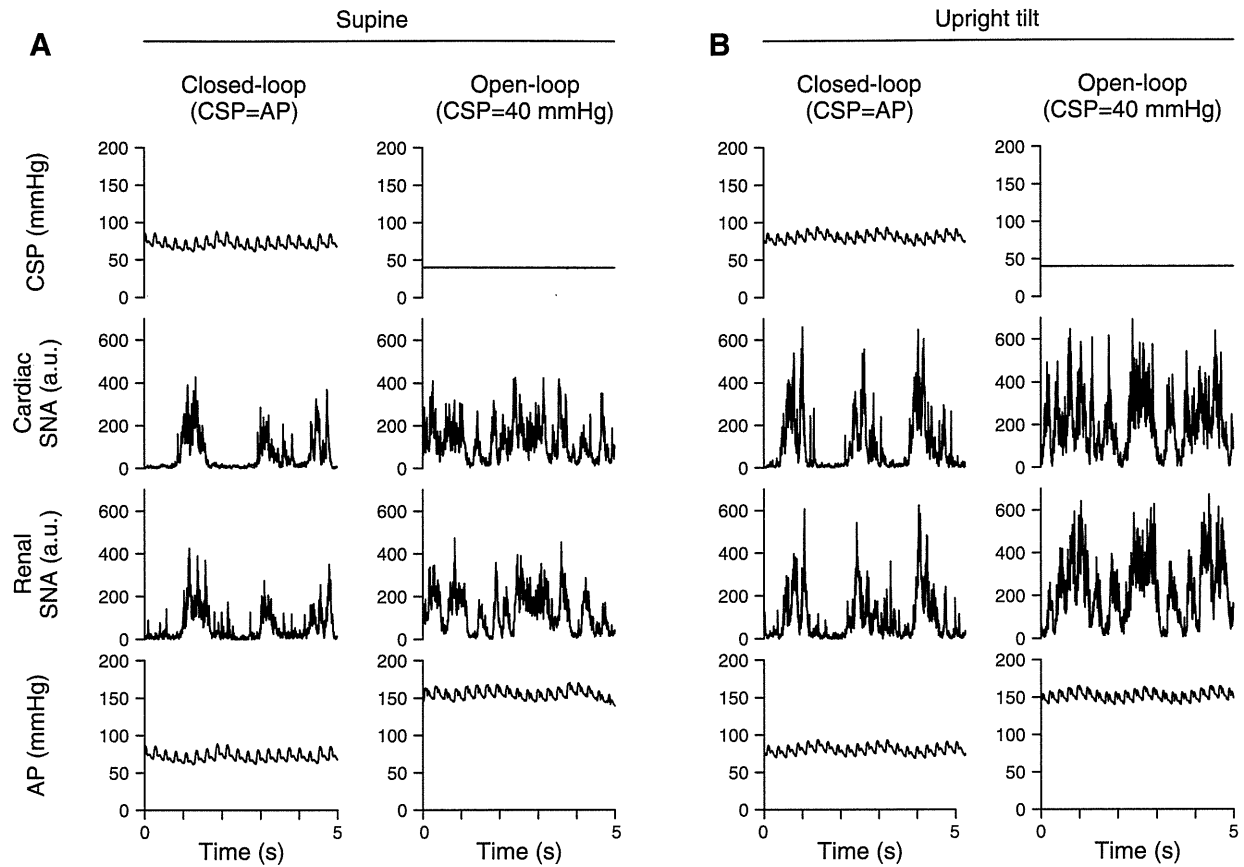


Fig. 9. Discharge characteristics of the renal and cardiac SNAs in supine (A) and 60° upright tilt postures (B) under the baroreflex closed-loop condition, where CSP was artificially matched with systemic AP, and under the open-loop condition, where CSP was fixed at 40 mmHg. Variables were resampled at 200 Hz. Data were obtained from the same animal studied in Fig. 1.

Table 2. Effect of upright tilt on parameters of baroreflex control of HR

	Supine	Upright tilt
P ₁ , beats/min	53 ± 11	67 ± 11*
P ₂ , beats · min ⁻¹ · mmHg ⁻¹	0.07 ± 0.03	0.07 ± 0.03
P ₃ , mmHg	93 ± 8	97 ± 12
P ₄ , beats/min	184 ± 17	191 ± 18
G _{max} , beats · min ⁻¹ · mmHg ⁻¹	-0.9 ± 0.2	-1.2 ± 0.4*

Values are means ± SD (n = 8) for the parameters of baroreflex control of heart rate (HR). *P < 0.05, supine vs. upright tilt.

Open-loop dynamic and static characteristics of the carotid sinus baroreflex in rats with chronic heart failure after myocardial infarction

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Abstract We estimated open-loop dynamic characteristics of the carotid sinus baroreflex in normal control rats and chronic heart failure (CHF) rats after myocardial infarction. First, the neural arc transfer function from carotid sinus pressure to splanchnic sympathetic nerve activity (SNA) and its corresponding step response were examined. Although the steady-state response was attenuated in CHF, the negative peak response and the time to peak did not change significantly, suggesting preserved neural arc dynamic characteristics. Next, the peripheral arc transfer function from SNA to arterial pressure (AP) and its corresponding step response were examined. The steady-state response and the initial slope were reduced in CHF, suggesting impaired end-organ responses. In a simulation study based on the dynamic and static characteristics, the percent recovery of AP was reduced progressively as the size of disturbance increased in CHF, suggesting that a reserve for AP buffering is lost in CHF despite relatively maintained baseline AP.

Keywords Systems analysis · Transfer function · White noise · Sympathetic nerve activity · Arterial pressure · Equilibrium diagram

Introduction

The arterial baroreflex is an important negative feedback system that stabilizes systemic arterial pressure (AP) against exogenous disturbances. The rapidness of AP regulation may be best described by the system dynamic characteristics. With respect to the sympathetic arterial baroreflex system, previous studies in rabbits [1] and rats [2] have indicated that the transfer function of the baroreflex neural arc from baroreceptor pressure input to efferent sympathetic nerve activity (SNA) exhibits “derivative” characteristics, which means that the dynamic gain of the SNA response to pressure perturbation becomes greater as the modulation frequency increases. On the other hand, the transfer function of the baroreflex peripheral arc from SNA to AP shows “low-pass” characteristics, which means that the dynamic gain of the AP response to SNA variation becomes smaller as the modulation frequency increases. In short, the neural arc provides an accelerating mechanism of the dynamic AP response in the arterial baroreflex system [1].

Although a number of studies have indicated that the baroreflex function is depressed in heart failure [3–6], dynamic characteristics of the arterial baroreflex in heart failure have not been fully described. The aim of the present study was to identify the open-loop dynamic characteristics of the carotid sinus baroreflex in a rat model of chronic heart failure (CHF) after myocardial infarction. To obtain a total picture of the AP regulation in CHF, we also estimated the open-loop static characteristics of the carotid sinus baroreflex.

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Materials and methods

Animals were cared for in strict accordance with the Guiding Principles for the Care and Use of Animals in the Field of Physiological Sciences, which has been approved by the Physiological Society of Japan. All experimental protocols were reviewed and approved by the Animal Subjects Committee at the National Cerebral and Cardiovascular Center.

Myocardial infarction

Coronary artery ligation was performed under sterile conditions in 8-week-old male Sprague-Dawley rats according to a previously established procedure [7]. After inducing anesthesia by halothane inhalation, the rat was intubated and mechanically ventilated. The left chest was opened at the fourth intercostal space and the left coronary artery was ligated with a 5-0 polypropylene suture (PROLENE, Ethicon, GA, USA). An electrocardiogram was monitored for 1 h after the coronary ligation, and the heart was defibrillated as necessary by manual prodding. Thereafter air was evacuated from the thoracic cavity and the incision was closed. The rats were allowed to recover from anesthesia, and were fed ad libitum with a standard laboratory chow and given free access to water. Although we initially planned to wait until the rats showed an objective sign of advanced heart failure such as facial edema or labored breathing, the rats with such severe heart failure were too weak to survive the acute baroreflex study described in the following section. Accordingly, the rats that survived for 100–200 days (156 ± 18 days) after myocardial infarction were used without solid criteria for starting the acute baroreflex study. Instead, the rats were regarded as experiencing CHF when the central venous pressure was higher than 2.5 mmHg or the biventricular weight was greater than 2.5 g/kg body weight at the time of the acute baroreflex study.

Acute baroreflex study

Experiments were performed in normal control ($n = 12$) and CHF ($n = 7$) rats. Sham operation was not performed in the control rats. Among the control rats, eight rats were matched with the CHF rats based on body weight, but they were younger in age due to the retardation of growth in the CHF rats. The remaining four rats were age-matched with the CHF rats, but they were heavier in body weight. Because none of the parameters of the baroreflex dynamic and static characteristics differed statistically between the two subgroups of the control rats, we report pooled data obtained from the 12 control rats.

Each rat was anesthetized with an intraperitoneal injection (2 ml/kg) of a mixture of urethane (250 mg/ml

and α -chloralose (40 mg/ml), and mechanically ventilated with oxygen-enriched room air. A venous catheter was inserted into the right femoral vein, and 20-fold diluted anesthetic mixture was administered continuously ($2\text{--}3 \text{ ml kg}^{-1} \text{ h}^{-1}$). An arterial catheter was inserted into the right femoral artery to measure AP. Heart rate (HR) was obtained from AP through a cardi tachometer. Another venous catheter was inserted into the left femoral vein and advanced into the inferior vena cava to measure central venous pressure and to supply Ringer solution ($6 \text{ ml kg}^{-1} \text{ h}^{-1}$).

A postganglionic branch from the splanchnic sympathetic nerve was exposed through a left flank incision, and a pair of stainless steel wire electrodes (Bioflex wire AS633, Cooner Wire, CA, USA) was attached to record SNA. The nerve and electrodes were covered with silicone glue (Kwik-Sil, World Precision Instruments, FL, USA) for insulation and fixation. To quantify the nerve activity, the preamplified nerve signal was band-pass filtered at 150–1,000 Hz, and then full-wave rectified and low-pass filtered with a cut-off frequency of 30 Hz. Pancuronium bromide ($0.4 \text{ mg kg}^{-1} \text{ h}^{-1}$) was administered to prevent muscular activity from contaminating the SNA recording. At the end of the experiment, we confirmed the disappearance of SNA in response to an intravenous bolus injection of a ganglionic blocker, hexamethonium bromide (60 mg kg^{-1}), and recorded the noise level.

Bilateral vagal and aortic depressor nerves were sectioned at the neck to avoid reflexes from the cardiopulmonary region and aortic arch. The carotid sinus regions were isolated from the systemic circulation bilaterally according to previously reported procedures [8, 9]. Briefly, a 7-0 polypropylene suture with a fine needle (PROLENE, Ethicon, GA, USA) was passed through the tissue between the external and internal carotid arteries, and the external carotid artery was ligated close to the carotid bifurcation. The internal carotid artery was embolized using two to three steel balls (0.8 mm in diameter, Tsubaki Nakashima, Nara, Japan) injected from the common carotid artery. Under these conditions, the brain stem area was perfused by patent bilateral vertebral arteries. The isolated carotid sinuses were filled with warmed Ringer solution through catheters inserted into the common carotid arteries. Carotid sinus pressure (CSP) was controlled using a servo-controlled piston pump. Heparin sodium (100 U kg^{-1}) was given intravenously to prevent blood coagulation. Body temperature was maintained at approximately 38°C with a heating pad.

Protocols

After the above surgical procedures were completed, reflex responses in SNA, AP, and HR to CSP input were

monitored for more than 30 min. The rat was excluded from further study and analysis in the event that the reflex responses became smaller within this period. Possible causes for the deterioration in the reflex responses include surgical damage to the carotid sinus nerves and brain ischemia due to the bilateral carotid occlusion.

To estimate the dynamic input-output relationship of the carotid sinus baroreflex, CSP was perturbed for 20 min using a Gaussian white noise (GWN) signal with the mean of 120 mmHg and standard deviation of 20 mmHg. The selection of the mean pressure and amplitude of GWN does not significantly affect the estimation of the system dynamic characteristics except for a factor of proportionality (see Appendix 1 for details). The switching interval of GWN was set at 500 ms. The input power spectral density was relatively constant up to 1 Hz, which covered the upper frequency range of interest with respect to the sympathetic arterial baroreflex in rats [2].

To estimate the static input-output relationship of the carotid sinus baroreflex, CSP was decreased to 60 mmHg for 4–6 min, and then increased stepwise from 60 to 180 mmHg in increments of 20 mmHg every minute [10].

Data analysis

Data were sampled at 200 Hz using a 16-bit analog-to-digital converter and stored on a dedicated laboratory computer system. In each rat, the noise level of SNA recorded after the administration of hexamethonium bromide was treated as zero. Because the absolute voltage of SNA varied among animals depending on the recording conditions, SNA averaged during the last 10 s at CSP of 60 mmHg in the stepwise input protocol was defined as 100%. The same normalization factor was used for the analysis of the baroreflex dynamic characteristics.

Dynamic characteristics of the baroreflex neural arc, peripheral arc, total baroreflex, and HR control were estimated by an open-loop transfer function analysis as follows [11]. Data were analyzed from 90 s after the initiation of the GWN input. The input-output data pairs were resampled at 10 Hz and segmented into 50% overlapping bins of 1,024 points each. For each segment, a linear trend was removed, and a Hanning window was applied. Fast Fourier transform was performed to obtain the frequency spectra of the input and output signals. The ensemble averages of the input power spectral density [$S_{XX}(f)$], output power spectral density [$S_{YY}(f)$], and cross spectral density between the input and output signals [$S_{YX}(f)$] were calculated over 12 segments, where f denotes frequency. Finally, the transfer function [$H(f)$] from input to output was estimated as:

$$H(f) = \frac{S_{YX}(f)}{S_{XX}(f)} \tag{1}$$

The transfer function is a complex-valued function that can be expressed by the modulus and phase at each frequency. In the present study, we refer to the modulus of the transfer function as the dynamic gain. To quantify the linear dependence between the input and output signals, a magnitude squared coherence function [$\text{Coh}(f)$] was calculated as:

$$\text{Coh}(f) = \frac{|S_{YX}(f)|^2}{S_{XX}(f)S_{YY}(f)} \tag{2}$$

The coherence function is a real-valued function ranging from zero to unity. When the output signal is perfectly explained by the linear dynamics with the input signal, the coherence value becomes unity. When the output signal is totally independent of the input signal, the coherence value becomes zero.

To facilitate understanding of the transfer function, the step response was also calculated as follows. The system impulse response was derived from the inverse Fourier transform of $H(f)$. The step response was then obtained from the time integral of the impulse response.

To quantify the open-loop static characteristics of the carotid sinus baroreflex, mean SNA, AP, and HR were obtained during the last 10 s at each CSP level of the stepwise input protocol. In each rat, data from two consecutive step cycles were averaged. The static characteristics of the baroreflex neural arc (the CSP-SNA relationship), the total baroreflex (the CSP-AP relationship), and the HR control (the CSP-HR relationship) were described by fitting four-parameter logistic functions to the input-output data as follows [10, 12]:

$$y = \frac{P_1}{1 + \exp[P_2(x - P_3)]} + P_4 \tag{3}$$

where x and y denote the input (CSP) and output (SNA, AP, or HR), respectively; P_1 is the response range of output; P_2 is the slope coefficient; P_3 is the midpoint pressure of input; and P_4 is the minimum value of output. For convenience, the maximum slope or the maximum gain of the logistic function is reported by a positive value as $P_1P_2/4$.

The static characteristics of the baroreflex peripheral arc (the SNA-AP relationship) were quantified by a linear regression analysis as follows [10, 13]:

$$\text{AP} = a \times \text{SNA} + b \tag{4}$$

where a and b represent the slope and intercept, respectively, of the regression line.

Statistical analysis

All data are presented in mean and SE values. Unpaired *t* tests were used to compare the parameters of the baroreflex dynamic and static characteristics between the control and CHF groups [13]. To compare the transfer functions between the two groups, we arbitrarily selected the dynamic gain values at 0.01, 0.1, and 1 Hz ($G_{0.01}$, $G_{0.1}$, and G_1). For the step response relating to the neural arc transfer function, the negative peak response (S_{peak}), the time to the negative peak (T_{peak}), the value of the step response at 10 s (S_{10}), the steady-state response at 50 s (S_{50}), and the ratio of the peak response to the steady-state response (S_{peak}/S_{50}) were calculated. For the step response relating to the peripheral arc, total baroreflex, or HR control, an initial slope of the response was calculated (see Appendix 2) in addition to S_{50} . Differences were considered significant when $P < 0.05$.

Simulation study

Once the open-loop dynamic and static characteristics of a system are both identified, closed-loop system responses can be simulated [14–16]. To compare closed-loop behavior of the carotid sinus baroreflex between control and CHF conditions, step inputs ranging from -10 to -60 mmHg were applied as exogenous disturbances, and resulting AP responses were simulated. Percent recovery was calculated as the magnitude of steady-state AP recovery relative to the size of step disturbance. The initial slope for the recovery response was also calculated (see Appendix 2).

Results

Postmortem examination confirmed that the left ventricular free wall was reduced to a membrane-like scar in the CHF group. Biventricular weights, both absolute and relative to body weight, were significantly greater in the CHF than in the control group (Table 1). The central venous pressure was significantly higher and the baseline AP and HR were

significantly lower in the CHF group. Although the duration after myocardial infarction ranged from 100 to 200 days in the CHF group, there was no significant correlation between the duration and biventricular weight ($y = 0.0012x + 2.53$, $r^2 = 0.035$, $P = 0.72$, x duration in days, y biventricular weight in grams per kilogram) or between the duration and central venous pressure ($y = -0.013x + 7.5$, $r^2 = 0.07$, $P = 0.58$, x duration in days, y central venous pressure in millimeters of mercury).

Typical experimental recordings obtained from a control rat are shown in Fig. 1a. m-SNA indicates a 2-s moving averaged signal of SNA. In the dynamic input protocol, CSP was changed dynamically according to a GWN signal. m-SNA varied dynamically in response to the CSP perturbation. Although AP changed dynamically, the AP variation seemed more sluggish than the SNA variation. Changes in HR were less obvious from the time series data. In the static input protocol, a stepwise increase in CSP decreased m-SNA, AP, and HR. The noise level of SNA obtained after the intravenous administration of hexamethonium bromide was set at zero. Because of the normalization procedure, m-SNA during the last 10 s at CSP of 60 mmHg approximated 100%. Figure 1b represents CSP and SNA signals sampled at 200 Hz during the dynamic input protocol. High CSP inputs suppressed SNA to a noise level.

Typical experimental recordings obtained from a CHF rat are shown in Fig. 2a. In the dynamic input protocol, CSP was changed dynamically according to a GWN signal. Although m-SNA varied dynamically in response to the CSP perturbation, changes in AP and HR were not obvious from the time series data. In the static input protocol, a stepwise increase in CSP decreased m-SNA, AP, and HR. The magnitudes of the responses in m-SNA, AP, and HR seem smaller than those in the control rat. Figure 2b represents CSP and SNA signals sampled at 200 Hz during the dynamic input protocol. High CSP inputs suppressed SNA to a noise level.

Dynamic characteristics of the carotid sinus baroreflex

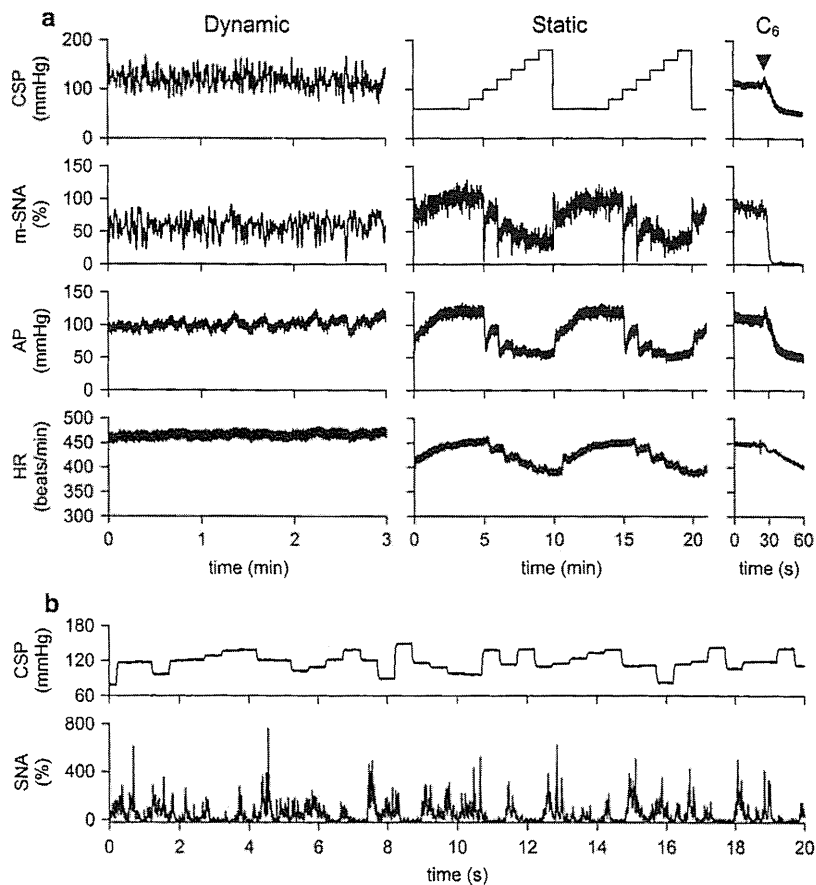
Figure 3 summarizes the open-loop dynamic characteristics of the carotid sinus baroreflex averaged for the control

Table 1 Age, body weight, biventricular weight, central venous pressure, mean arterial pressure, and heart rate of the normal control and chronic heart failure (CHF) rats

Data are presented as mean \pm SE
 ** $P < 0.01$ and * $P < 0.05$ by unpaired *t* test

	Control ($n = 12$)	CHF ($n = 7$)	<i>P</i> value
Age at experiment (weeks)	24 \pm 3	30 \pm 3	0.237
Body weight (g)	565 \pm 28	538 \pm 19	0.474
Biventricular weight (g)	1.16 \pm 0.04	1.45 \pm 0.08**	0.002
Biventricular weight (g kg body weight ⁻¹)	2.05 \pm 0.06	2.71 \pm 0.13**	<0.001
Central venous pressure (mmHg)	2.0 \pm 0.2	5.4 \pm 0.9**	<0.001
Mean arterial pressure (mmHg)	134 \pm 4	121 \pm 4*	0.037
Heart rate (beats min ⁻¹)	414 \pm 11	350 \pm 12**	0.001

Fig. 1 **a** Typical recordings (10-Hz decimated data) of carotid sinus pressure (CSP), 2-s moving averaged sympathetic nerve activity (m-SNA), arterial pressure (AP), and heart rate (HR) obtained from a control rat. In the dynamic protocol, CSP was changed according to a Gaussian white noise signal. In the static protocol, CSP was increased from 60 to 180 mmHg. Hexamethonium bromide (C_6) was administered intravenously at the end of the experiment (arrowheads). The noise level of the nerve activity recorded after C_6 administration was assigned 0%, while the m-SNA value averaged for the last 10 s at CSP of 60 mmHg was assigned 100%. **b** CSP and SNA sampled at 200 Hz during the dynamic input protocol



and CHF rats. The neural arc transfer function from CSP to SNA showed derivative characteristics (Fig. 3a). $G_{0.01}$ tended to be lower in the CHF rats, whereas $G_{0.1}$ and G_1 did not differ significantly between the two groups (Table 2). The phase plot showed an out-of-phase relationship in the frequency range from 0.01 to 1 Hz in both groups. The coherence plot with values less than unity suggests that SNA contained an unknown noise signal unrelated to the baroreflex and/or a nonlinear system response to the CSP input. The bottom panel of Fig. 3a represents the step responses of SNA for a unit increase in CSP. Although S_{10} and S_{50} were significantly attenuated in the CHF rats, S_{peak} , T_{peak} , and S_{peak}/S_{50} did not differ significantly between the two groups (Table 2).

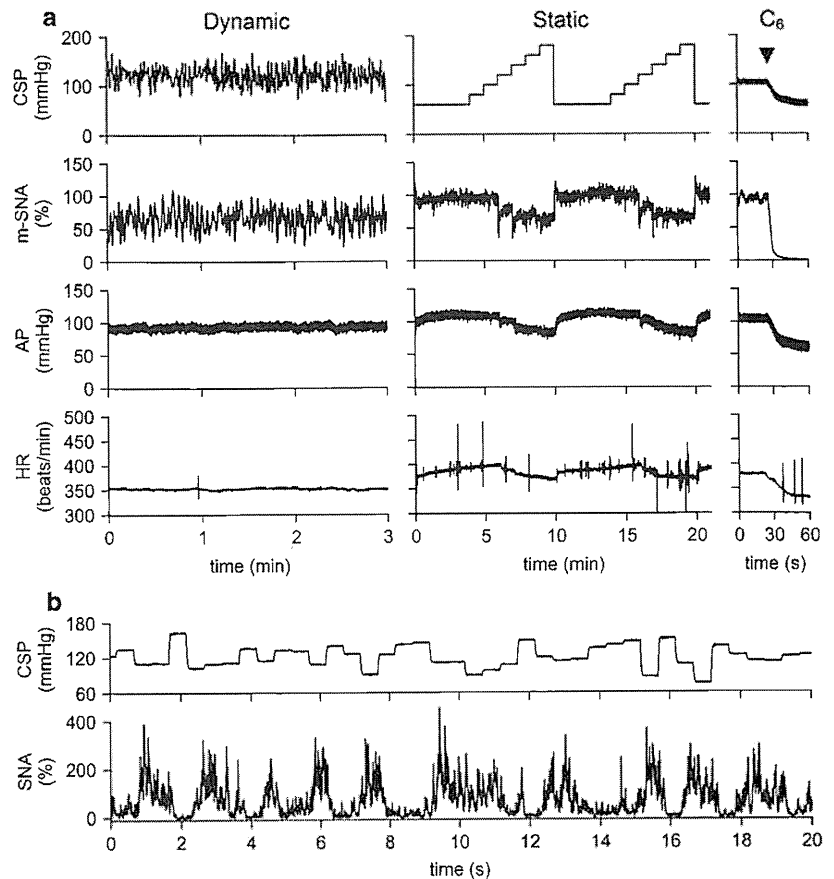
The peripheral arc transfer function from SNA to AP showed low-pass characteristics (Fig. 3b). $G_{0.01}$ and $G_{0.1}$ were significantly smaller in the CHF rats, whereas G_1 did not differ significantly between the two groups (Table 2). The phase was close to zero radians at the lowest frequency and delayed with an increase in frequency in both groups. The coherence plot with values less than unity indicates that a part of AP variation was not explained by the linear dynamics with the SNA variation. In the step response of AP for a unit increase in SNA (Fig. 3b, bottom), S_{50} was

significantly smaller and the initial slope (the dotted line) was significantly gentler in the CHF than in the control rats (Table 2).

The total baroreflex transfer function from CSP to AP showed low-pass characteristics (Fig. 3c). $G_{0.01}$ and $G_{0.1}$ were significantly smaller in the CHF than in the control rats (Table 2), but G_1 did not differ significantly. The phase was close to $-\pi$ radians at the lowest frequency in both groups, reflecting the negative feedback operation attained by the total baroreflex. The phase delayed with an increase in frequency. The coherence values seem lower than those in the peripheral arc transfer function. In the step response of AP for a unit increase in CSP (Fig. 3c, bottom), both S_{50} and the initial slope (the dotted line) were significantly attenuated in the CHF in comparison with the control rats (Table 2).

The transfer function from CSP to HR also displayed low-pass characteristics (Fig. 3d). $G_{0.01}$ and $G_{0.1}$ were significantly smaller in the CHF than in the control rats (Table 2). We did not compare G_1 because the coherence values close to zero and the irregular change in the phase shift above 0.8 Hz suggested poor reliability of the estimated transfer function. In the step response of HR for a unit increase in CSP (Fig. 3d, bottom), both S_{50} and the

Fig. 2 **a** Typical recordings (10-Hz decimated data) of CSP, m-SNA, AP, and HR obtained from a chronic heart failure rat. In the dynamic protocol, CSP was changed according to a Gaussian white noise signal. In the static protocol, CSP was increased from 60 to 180 mmHg. The noise level of the nerve activity recorded after C_6 administration was assigned 0%, while the m-SNA value averaged for the last 10 s at CSP of 60 mmHg was assigned 100%. **b** CSP and SNA sampled at 200 Hz during the dynamic input protocol



initial slope (the dotted line) were significantly attenuated in the CHF in comparison with the control rats (Table 2).

Static characteristics of the carotid sinus baroreflex

Figure 4 summarizes the open-loop static characteristics of the carotid sinus baroreflex obtained from the control and CHF rats. The baroreflex neural arc showed a decreasing SNA response with an increase in CSP (Fig. 4a). The response range of SNA was significantly narrower in the CHF rats (Table 3). Consequently, the minimum SNA was significantly higher in the CHF rats. The midpoint pressure on the CSP axis was significantly lower in the CHF rats. Despite the significant attenuation in the response range of SNA, the maximum slope of the neural arc was not reduced in the CHF rats compared with the control rats.

The peripheral arc from SNA to AP approximated a straight line in both the control and CHF rats (Fig. 4b). The slope of the regression line was significantly less steep in the CHF rats, whereas the AP intercept did not differ significantly between the two groups (Table 3).

The total baroreflex function from CSP to AP approximated an inverse sigmoidal curve (Fig. 4c). The response range of AP was significantly narrower in the CHF rats

(Table 3). The slope coefficient, the midpoint pressure on the CSP axis, and the minimum AP did not differ significantly between the two groups. The maximum gain was significantly smaller in the CHF rats compared with the control rats.

The static CSP-HR relationship also approximated an inverse sigmoidal curve (Fig. 4d). The response range of HR was significantly narrower, and the minimum HR was significantly lower in the CHF rats (Table 3). The midpoint pressure on the CSP axis was significantly lower in the CHF rats. The slope coefficient and the maximum slope did not differ significantly between the two groups.

The baroreflex equilibrium diagram is obtained by plotting the neural and peripheral arcs on a pressure-SNA plane (Fig. 4e). The ordinate is either CSP (for the neural arc) or AP (for the peripheral arc). The intersection between the neural and peripheral arcs gives the closed-loop operating point [17, 18]. The operating-point AP (the horizontal arrow) was significantly lower in the CHF rats, whereas the operating-point SNA (the vertical arrow) did not differ significantly between the two groups (Table 3). The total baroreflex gain at the operating point, calculated from the product of the tangential slope of the neural arc ($\Delta SNA/\Delta CSP$) and the slope of the peripheral arc ($\Delta AP/$