Selective impairment of central mediation of baroreflex in anesthetized young adult Fischer 344 rats after chronic intermittent hypoxia

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OBSTRUCTIVE SLEEP APNEA (OSA) is associated with cardiovascular complications and substantial morbidity (11, 44, 49, 62). In OSA patients, baroreflex control of heart rate (HR) (baroreflex sensitivity) is reduced (3, 4, 38, 42). Attenuation of baroreflex sensitivity is closely associated with several clinical conditions including heart failure (16, 31, 53) and is considered an independent risk factor for sudden death (26). Therefore, improved understanding of chronic intermittent hypoxia (CIH)-associated changes of baroreflex function is clearly needed for improved formulation of interventional strategies aimed at reducing the morbidity and mortality associated with OSA.

CIH during sleep, one of the characteristics of OSA, has been used as a useful model for OSA (20). As in OSA, baroreflex control of HR is significantly reduced after CIH in Sprague-Dawley (SD) rats (32). Consistent with these findings, we observed (Ref. 35; Lin M, Gozal D, Liu R, Cheng Z, unpublished observations) that CIH exposure led to altered baroreflex function and remodeling of vagal efferent axon projections to cardiac ganglia in C57BL/6J mice. In addition, we found that postnatal CIH exposure led to altered baroreflex function in adult rats and reduced vagal efferent axon projections to cardiac ganglia (54). Furthermore, early postnatal CIH exposure leads to long-term substantial reductions in vagal afferent projections to the nucleus of the solitary tract (NTS) and significant increases in the total number of nucleus ambiguus (NA) motoneurons (47).

Notwithstanding advances in our understanding of the baroreceptor reflex impairment following CIH in recent years (Refs. 32, 34, 36, 47, 48, 54, 55), very little is currently known about the functional changes and associated remodeling of baroreceptor afferent, central, and efferent components of the reflex. In the present study, we hypothesized that CIH impairs the functions of baroreceptor afferent, central, and efferent components of the baroreflex circuitry in Fischer 344 (F344) rats. We aimed to determine which and how neural components within the baroreflex circuitry were changed by CIH.

MATERIALS AND METHODS

F344 rats (3–4 mo of age) were used. Procedures were approved by the University of Central Florida Animal Care and Use Committee and followed the guidelines established by the National Institutes of Health. Efforts were made to minimize the number of animals used.

Intermittent Hypoxia Exposure

Animals were housed in Plexiglas chambers (30 × 20 × 20 in.3; OxyCycler model A44XO, BioSpherix Instruments, Redfield, NY) in a room in which light and dark cycles were set at 12:12 h (6:00 AM to 6:00 PM). O2 concentration in these chambers was continuously measured by an O2 analyzer and was controlled through a gas valve outlet by a computerized system. O2 concentrations in the chambers...
were programmed and adjusted automatically. Any deviation from the desired O₂ concentration was corrected by adding pure N₂ or O₂ through solenoid valves. Ambient CO₂ in the chambers was periodically monitored and maintained at 0.03% by adjusting overall chamber ventilation. Humidity was measured and maintained at 40–50%. Temperature was kept at 22–24°C. The intermittent hypoxia (CIH) profile consisted of alternating 21% (90 s) and 10% (90 s) O₂ every 6 min for 12 h during the light cycle and O₂ maintained at 21% for the night period, with an overall exposure duration of 35–50 days. The room air (RA) control animals were housed in room air under the same conditions as CIH-exposed animals, except that the concentration of O₂ was maintained at 21% throughout the duration of exposure.

Surgical Procedures

Rats were anesthetized and ventilated with isoflurane (2%) in 95% O₂-5% CO₂ through the trachea. Body temperature was maintained at 37 ± 1°C with a homeostatic blanket (Harvard) and a rectal probe. Polyethylene catheters (PE-50) were placed in the left femoral artery to monitor arterial pressure (AP) and in both left and right femoral veins to infuse drugs [sodium nitroprusside (SNP), phenylephrine (PE), anesthetic agents]. The hind paw pinch withdrawal reflex was used to assess the level of anesthesia.

Baroreflex Control of HR and Baroreceptor Afferent Function in Isoflurane-Anesthetized Rats

Baroreflex sensitivity. Baseline values of mean arterial blood pressure (MAP) and HR and the MAP and chronotropic responses to sequential SNP-PE applications were measured. Arterial blood pressure was measured with a PowerLab Data Acquisition System (PowerLab/8 SP) that was connected to a pressure transducer (iWorx/CB Sciences, BP-100). HR was calculated from pulse pressure waves with the ratemeter function of Chart 5.2 software provided in the PowerLab System. SNP and PE (Sigma, St. Louis, MO) were freshly prepared, diluted in 0.9% NaCl, and administered intravenously by sequential bolus injections. SNP (30–60 μg/kg in 50–100 μl saline) was injected first, and after 30–60 s PE (150–1,500 μg/kg in 50–200 μl saline) was then injected. These doses of SNP-PE induced a fast and large depressor response followed by an increase in MAP, such that the full range of the baroreceptor function curve could be fitted with the sigmoid logistic function (37). Manipulations of MAP were completed within 2 min to limit the extent of baroreceptor resetting. The doses of SNP and PE were selected to produce similar changes in arterial pressure in RA and CIH rats. Baseline values of MAP and HR were averaged from a 30-s interval before SNP injection. After SNP-PE injections, MAP and HR returned to baseline values. HR responses to MAP changes induced by sequential administration of SNP and PE included two phases: tachycardic and bradycardic responses. During the tachycardic (SNP) phase, MAP and HR changes were measured in the time window as indicated by the hatched boxes shown in Fig. 1, i.e., the baseline to the nadir of MAP. During the bradycardic (PE) phase, MAP and HR changes were measured in the time window as indicated by the hatched boxes in Fig. 1, i.e., from the peak of HR to the nadir of HR. MAP and corresponding HR were sampled and averaged every second. We performed separate linear regression analyses of the HR change (ΔHR)-MAP change (ΔMAP) relationships for responses to SNP and PE in each animal. The slope of the regression line was used as an index of baroreflex sensitivity. The correlation coefficient r² was used to determine the goodness of fit. In our analyses, r² was >0.81 for each of the animals used.

Baroreceptor afferent function. The left aortic depressor nerve (ADN) was identified in the cervical region with a dissecting microscope. The left ADN was carefully isolated from surrounding connective tissue with fine glass tools to avoid injury to the nerve. The left ADN was then placed on a bipolar platinum electrode (0.12-mm outer diameter). The ADN nerve and electrode were covered with mineral oil. ADN activity (ADNA) was amplified (×10,000) with the band-pass filters set between 300 and 1,000 Hz by an AC amplifier (model 1800, A-M Systems). ADNA, integrated ADNA (Int ADNA), phasic arterial pressure (PAP), HR, ECG, and body temperature were all recorded and simultaneously displayed on different channels of the PowerLab System. Chart 5.2 software and Sigma Plot 9.0 were used for data acquisition and analysis. The signal-to-noise ratio for ADNA was >6:1 in all experiments.
The ADNA signal occurred as rhythmic bursts that were synchronized with the arterial pulse pressure (PAP; Fig. 2). The ADNA signal was integrated with a 10-ms time constant to obtain the Int ADNA. The “ADNA silent” intervals or the “noise level” between the ADNA bursts is shown in the small boxes in the Int ADNA signal of Fig. 2. The averaged value of 30 ADNA silent intervals was used to determine the noise level for Int ADNA. This averaged noise level was subtracted from the original Int ADNA signal to obtain the corrected Int ADNA. The corrected Int ADNA and MAP were used to construct a baroreceptor function curve. For simplicity, we use “Int ADNA” to indicate corrected Int ADNA in the text below. The baroreceptor function curve was calculated from data measured during the rising phase of the PE-induced AP change starting from the nadir of the SNP-induced fall in AP to the maximum of the AP. QRS waves of the ECG signal were used to automatically define cardiac cycles by the Chart 5.2 Macro function (arrows in Fig. 2). The baroreceptor function curve was fitted by plotting the percent change of the mean Int ADNA per cardiac cycle relative to the Int ADNA baseline value before drug administration against MAP, using a sigmoid logistic function (28, 37). The logistic function for Int ADNA used the mathematical expression: 

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

where \( X \) = mean arterial pressure, \( Y \) = Int ADNA (% baseline), \( P_1 \) = maximum – minimum Int ADNA (range), \( P_2 \) = slope coefficient, \( P_3 \) = mean arterial pressure at 50% of the Int ADNA range (Pmid), and \( P_4 \) = maximum Int ADNA. \( P_3 \) and \( P_4 \) were calculated from the third derivative of the logistic function, and they were expressed as \( P_3 = P_1 - (1.317/P_2) \) and \( P_4 = P_1 + (1.317/P_2) \). The maximum gain or slope (Gmax) was calculated at Pmid from the first derivative of the logistic function (Gmax = \(-P_3 \times P_2/P_4\)). Approximately 100–300 data points measured over 60–120 s were used to construct a baroreceptor function curve with Sigma Plot software. The correlation coefficient \( r^2 \) was used to determine the goodness of fit. In our analysis, \( r^2 \) was >0.95 for each animal used. Parameters of the baroreflex function curve were averaged within groups.

**HR and Blood Pressure Responses to Electrical Stimulation of Left ADN in Ketamine-Acetpromazine-Anesthetized Rats**

Separate groups of rats were instrumented as described above, i.e., the left ADN was identified, isolated, and placed on a bipolar platinum electrode (0.12-mm outer diameter). The left ADN was then crushed at a point caudal to the electrode. The nerve was stimulated with rectangular current pulses (50 \( \mu \)A, 1 ms) that were delivered to the electrode at frequencies of 4, 8, 16, and 32 Hz from a Grass S48 Stimulator (Grass Instruments, West Warwick, RI) through an isolation unit (Grass model PSIU 6). The duration of the stimulus train was 20 s, with 5 min allowed for recovery between periods of stimulation. Responses to each frequency of stimulation were measured at least twice in each experiment, with the order of changes of frequency presentation reversed during the second round of stimulation. The responses were repeatable. The maximal changes in HR and MAP during ADN stimulation were measured. In addition, the time course of the HR responses calculated both as percentage of the maximum response and as percentage of baseline was assessed. The stimulation-induced changes in MAP and HR were abolished after the left ADN was crushed cranial to the electrode, confirming that the responses were reflex in nature.

**HR and Blood Pressure Responses to Electrical Stimulation of Right Vagus Nerve in Ketamine-Acetpromazine-Anesthetized Rats**

The right cervical vagus nerve was isolated from surrounding connective tissue with fine glass tools and sectioned. The distal cut end of the vagus nerve was placed on a bipolar platinum electrode and stimulated electrically with rectangular current pulses (500 \( \mu \)A, 1 ms) at frequencies of 1, 5, 10, 20, and 30 Hz. The data were analyzed as described for ADN stimulation. The stimulation-induced changes in HR and AP were abolished after the vagus nerve was crushed caudal to the electrode, confirming that the responses were indeed due to vagal efferent activity.

**Experimental Protocols**

After placement of the arterial and venous catheters, the rats were allowed to stabilize for a period of 30 min before the actual experiments began. Three experiments were performed. In experiment 1, baroreflex control of HR and baroreceptor afferent function were assessed under isoflurane anesthesia (2%). Seagard et al. (52) have reported that isoflurane does not depress the baroreflex at concentrations <2.6%. In experiment 2, HR and MAP responses to electrical stimulation of the left ADN were measured in rats anesthetized with ketamine (18 mg·kg\(^{-1}\)·h\(^{-1}\) iv) and acepromazine (0.36 mg·kg\(^{-1}\)·h\(^{-1}\) iv) delivered via a microinfusion pump. Ma et al. (37) previously demonstrated that reflex decreases in HR in response to ADN stimulation with this anesthetic regimen were largely preserved. After 1 h of ketamine-acepromazine, when HR and MAP had reached and stabilized at the new baseline levels, HR and MAP responses to left ADN stimulation were measured. In experiment 3, MAP and HR responses to stimulation of the right vagus nerve were measured.
Baseline Values of Arterial Blood Pressure and HR

Under isoflurane anesthesia, baseline values for MAP and HR of RA and CIH rats were comparable \( [n = 19 \text{ (RA)}, 16 \text{ (CIH); } P > 0.10] \) (Table 1). Under ketamine and acepromazine, baseline values for MAP and HR of RA and CIH rats were not significantly different \( [n = 8 \text{ (RA)}, 9 \text{ (CIH}; P > 0.10] \). MAP and HR were significantly increased in ketamine-acepromazine-anesthetized rats compared with MAP and HR in corresponding isoflurane-anesthetized (RA and CIH) groups \( (P < 0.05; \text{Table 1}) \). Since the baselines of MAP and HR were comparable in RA and CIH groups for either of the anesthetic agents, this allowed us to examine ADNA changes in response to blood pressure changes and HR responses to left ADN and vagal efferent stimulation starting from the same baseline levels in RA and CIH rats.

Baroreflex Control of HR (Baroreflex Sensitivity)

HR responses to MAP changes induced by sequential SNP-PE administration included two phases: tachycardic and bradycardic responses. SNP application induced hypotension that reached a similar nadir in RA and CIH rats \( (\text{RA } 46 \pm 2 \text{ mmHg vs. CIH } 45 \pm 1 \text{ mmHg}; P > 0.10) \). Subsequent injection of PE produced a ramp increase in MAP that reached a similar maximum of 180 \pm 4 \text{ (RA)} and 173 \pm 5 \text{ (CIH) mmHg} \ (P > 0.10). Therefore, SNP-induced hypotension and PE-induced hypertension were similar in RA and CIH rats. This allowed us to examine baroreflex sensitivity over a similar range of blood pressure changes in RA and CIH rats.

For baroreflex sensitivity, \( \Delta \text{MAP} \) and \( \Delta \text{HR} \) were measured as shown in Fig. 3. In the tachycardic and bradycardic phases, data were fitted as separate regression lines in representative RA and CIH rats (Fig. 3A, top and bottom, respectively). The average slopes of the regression lines were significantly different for both tachycardic and bradycardic phases: \(-0.69 \pm 0.30 \text{ (RA) vs. } -0.22 \pm 0.02 \text{ (CIH) beats per minute (bpm)/mmHg} \ (P < 0.05, n = 8; \text{Fig. 3B, top}) \) and \(-1.35 \pm 0.12 \text{ (RA) vs. } -0.83 \pm 0.18 \text{ (CIH)} \ (P < 0.05, n = 8/\text{group}) \).
maximal Int ADNA (Table 2), indicating that the ADNA response to MAP increase was a larger percentage of baseline activity. The values of $P_1$, $P_2$, and $P_{sat}$ for the baroreceptor afferent function curve did not differ significantly between groups. With the group means of the parameters in the logistic function, the logistic function curves were reconstructed and were significantly different in RA vs. CIH rats (Fig. 4C).

Central Mediation of Baroreflex Control of HR and MAP

Electrical stimulation of the left ADN evoked frequency-dependent decreases in HR and MAP in both RA and CIH rats (Fig. 5A). CIH significantly decreased the peak bradycardic and depressor responses to stimulation (Fig. 5B). Furthermore, at all frequencies of stimulation the fall in HR developed more slowly in CIH rats than in RA rats (Fig. 5A). Group data compiled for stimulation at a frequency of 16 Hz show that CIH significantly slowed the HR response (Fig. 5C) and reduced the magnitude of the decrease in HR after stimulus onset (Fig. 5D; ANOVA, $P < 0.05$).

Vagal Efferent Control of HR

Electrical stimulation of the right vagal efferent nerve evoked frequency-dependent decreases in HR and MAP in both RA and CIH rats (Fig. 6A). Interestingly, CIH significantly increased the peak magnitude of bradycardia at low frequencies of stimulation (1–10 Hz) (Fig. 6B). The depressor response to 10-Hz stimulation was also enhanced in CIH rats (Fig. 6B). At higher frequencies of stimulation, HR and AP responses were not significantly different between groups. As shown in Fig. 6A, the fall in HR occurred more rapidly in CIH rats compared with RA rats. The group data for the HR response to 10 Hz show that CIH significantly increased the rate of the fall in HR (Fig. 6C) and increased the magnitude of the HR response at any given time after stimulus onset (Fig. 6D; ANOVA, $P < 0.05$).

Table 2. Parameters defining the baroreceptor afferent (ADNA % baseline) function curve in RA and CIH rats

<table>
<thead>
<tr>
<th></th>
<th>$r^2$</th>
<th>$P_1$ (%)</th>
<th>$P_2$ (gain coefficient)</th>
<th>$P_3$, mmHg</th>
<th>$P_4$, %</th>
<th>$G_{max}$, %/mmHg</th>
<th>$P_{th}$, mmHg</th>
<th>$P_{sat}$, mmHg</th>
</tr>
</thead>
<tbody>
<tr>
<td>RA ($n = 6$)</td>
<td>0.97±0.005</td>
<td>−258.76±37.29</td>
<td>0.06±0.007</td>
<td>123.41±3.67</td>
<td>260.88±30.42</td>
<td>3.80±0.49</td>
<td>100.48±5.36</td>
<td>146.34±3.42</td>
</tr>
<tr>
<td>CIH ($n = 8$)</td>
<td>0.97±0.003</td>
<td>−354.46±15.02</td>
<td>0.06±0.003</td>
<td>122.32±2.06</td>
<td>358.51±14.96</td>
<td>5.05±0.28</td>
<td>98.95±1.69</td>
<td>145.68±2.87</td>
</tr>
<tr>
<td>$P$ value</td>
<td>NS</td>
<td>&lt;0.05</td>
<td>NS</td>
<td>NS</td>
<td>&lt;0.01</td>
<td>&lt;0.05</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

Values are means ± SE for $n$ rats. ADNA, aortic depressor nerve activity; $r^2$, correlation coefficient; $P_1$, maximum − minimum integrated ADNA (Int ADNA) (range); $P_2$, slope coefficient; $P_3$, MAP at 50% of Int ADNA range; $P_4$, maximum Int ADNA; $G_{max}$, maximum baroreceptor gain or slope; $P_{th}$, $P_3 - (1.317/P_2)$; $P_{sat}$, $P_3 + (1.317/P_2)$; NS, not significant.
DISCUSSION

CIH significantly reduced baroreflex control of HR in isoflurane-anesthetized F344 rats. We hypothesized that CIH would decrease the function of afferent, central, and vagal efferent components of the baroreflex, and that the total effect of all these components could be additive and thereby contribute to a global attenuation of baroreflex control of HR. Contrary to this hypothesis, we found that CIH caused differential effects on the three components. While the overall central mediation of HR control was significantly impaired, CIH enhanced baroreceptor afferent function and augmented the HR response to vagal efferent stimulation. These findings suggest that the decreased baroreflex sensitivity following CIH is not caused by changes in peripheral components of the reflex arc. Instead, functional deficits of the central nervous system appear to be responsible.

Blood Pressure and HR in Anesthetized F344 Rats

Fletcher (13–15) reported that CIH (8 h/day for 35 days) induced systemic hypertension by 13 mmHg in rats. This observation has been subsequently confirmed by other laboratories (cf. Ref. 63). Recently, data in the mouse model confirmed this finding. We found a 20% increase in arterial blood pressure and a 12% increase in HR in CIH-exposed mice (36). The concurrence of hypertension and tachycardia may represent a sympathovagal imbalance, as has been observed in patients with OSA and experimental models involving exposure to CIH (14).

In contrast to SD rats and C57BL/6J mice, CIH did not significantly alter MAP and HR in F344 rats when assessed under isoflurane or ketamine-acepromazine anesthesia. Differences in the duration or timing of CIH might have contributed to differences in pressor responses. However, the CIH profile...
that we used was similar to that used in other studies in which hypertension was observed (23). A more likely possibility is that the anesthetic agents isoflurane and ketamine-acepromazine suppressed CIH-induced hypertension and tachycardia. Telemetric recording of blood pressure should be used in future studies to test whether CIH induces hypertension in conscious F344 rats. A third possibility is that F344 rats may be resistant to CIH-induced hypertension. The F344 rat has been shown to be resistant to aging-associated increases in AP, obesity, and atherosclerosis (9).

**Impairment in Baroreflex Control of HR**

Previous studies have demonstrated that CIH significantly impairs baroreflex control of HR in conscious SD rats and Avertin (tribromoethanol-amyl alcohol)-anesthetized and conscious C57BL/6J mice (35, 36). Our data confirm that the baroreflex is also impaired in isoflurane-anesthetized F344 rats. Obviously, any anesthetic can affect baroreflex sensitivity. Indeed, ketamine-acepromazine is a blocker of glutamatergic transmission through \( N \)-methyl-\( d \)-aspartate (NMDA) receptors, and these receptors play a major role in central baroreceptor processing, likely in the NTS and NA (25, 41). Therefore, the baroreflex sensitivity in isoflurane-anesthetized rats, or HR and MAP responses to left ADN stimulation in ketamine-acepromazine-anesthetized rats, was expected to be lower than that in conscious animals.

A limitation of the present study is that we could not distinguish whether CIH selectively impaired the parasympathetic vs. sympathetic limbs of the baroreflex. We purposely chose to measure HR responses to relatively rapid changes in blood pressure evoked by sequential bolus injections of SNP and PE in an attempt to involve parasympathetic mechanisms. Using selective autonomic receptor blockers, previous studies in our lab (8) and by others (24, 56) indicate that the bradycardic responses to abrupt increases in blood pressure, as occur during bolus injections of PE, are mediated primarily by...
parasympathetic activation. Similarly, parasympathetic activation is mainly responsible for the baroreflex-mediated decrease in HR during ADN stimulation (10). Therefore, it is likely that impaired parasympathetic control contributes to decreased baroreflex sensitivity in CIH rats. Future studies utilizing selective autonomic receptor blockers are needed to define the contributions of parasympathetic and sympathetic defects to the impaired baroreflex control of HR in CIH rats.

The SNP-induced hypotension preceding the PE-induced increase in blood pressure may potentially cause baroreflex resetting and/or reflex increases in vasopressin (5). The rapidity of the pressure changes and the expected high baseline levels of vasopressin in the anesthetized rat make it unlikely that these changes would influence our results. In addition, CIH uniformly impaired baroreflex responses to SNP- and PE-induced changes in blood pressure, as well as to ADN stimulation. Therefore, it is assumed that even if there were an arginine vasopressin release during hypotension, it would not dramatically reset the baroreflex since the ramp change in blood pressure was completed within 2 min in both RA and CIH rats (17, 58).

**Augmented Baroreceptor Afferent Sensitivity**

Initially we suspected that decreased arterial compliance, as reported in patients with OSA and animals exposed to CIH (27, 45), would limit vascular distension of aortic arch and carotid sinuses and consequently decrease baroreceptor sensitivity. To our surprise, baroreceptor sensitivity, as derived from recordings of ADNA, was significantly increased in CIH rats. The mechanism of this effect is unclear. Increased baroreceptor sensitivity accompanied by decreased vascular compliance (27, 45) suggests an increase in sensitivity of the baroreceptor nerve terminals to stretch, possibly mediated by an intrinsic change in membrane properties and/or sensitization by neurohumoral factors such as epinephrine (30, 57). In the present study, we recorded baroreceptor activity in the ADN. Whether CIH similarly increases carotid sinus baroreceptor sensitivity remains to be determined.

Interestingly, CIH increases carotid body chemoreceptor sensitivity to hypoxia, leading to reflex increases in respiration and efferent sympathetic nerve activity (46). After CIH, the increase in chemoreceptor sensitivity persists even after the hypoxic stimulus is removed (46). The sustained chemoreceptor discharge produces a lasting increase in baseline sympathetic nerve activity. Whether the increases in chemoreceptor activity and central respiratory drive contribute to decreased baroreflex control of HR via central interactions between these reflexes should be elucidated in the future.

**Augmented HR Responses to Vagal Efferent Stimulation**

Vagal efferent nerves comprise the parasympathetic output of the baroreflex arc. We hypothesized that CIH would attenuate the vagal efferent control of HR. Our present data showed that, contrary to our original hypothesis, the peak amplitude of the HR response to vagal stimulation was significantly increased and the time from stimulus onset to the peak amplitude of HR responses was significantly shortened at low stimulation frequencies, indicating that vagal efferent control of HR was augmented after CIH. With higher-frequency vagal stimulation, however, the HR responses were similar in RA and CIH groups. Therefore, impairment of baroreflex control of HR in CIH rats cannot be attributed to changes in vagal efferent function and/or chronotropic responsiveness of the heart. These data are consistent with our recent data (36) in which HR responses to vagal efferent stimulation were increased at low frequencies of stimulation in mice exposed to CIH. Such an increase of parasympathetic cardiac efferent control may be associated with remodeling of NA motoneuron projections to the cardiac ganglia (Refs. 1, 6, 7; Lin M, Gozal D, Liu R, Cheng Z, unpublished observations).

**Attenuated Central Mediation of Baroreflex Control of HR**

Consistent with our original hypothesis, the present data show that the HR responses to ADN stimulation are significantly decreased in CIH rats. These data, together with our other findings, suggest that dysfunction of central neural pathways is the major cause of baroreflex impairment in CIH rats.

To minimize the extent of surgical manipulation and the duration of experiments, the remaining buffer nerves (right ADN and carotid sinus nerves) were left intact. Therefore, it is possible that intact baroreceptor fibers in those nerves may have buffered the reflex fall in blood pressure and reduced the magnitude of bradycardia during stimulation of the left ADN. Given the reduced central baroreflex function found in CIH rats, it is reasonable to predict that alternative reflex pathways would provide less effective buffering of responses to ADN stimulation in CIH rats. Consequently, denervation of the remaining buffer nerves would be expected to enhance responses to ADN stimulation to a greater extent in RA rats than in CIH rats. Such an effect would increase the magnitude of the difference in reflex response between RA and CIH rats rather than reduce it. Furthermore, a previous study has shown that denervation of the contralateral ADN and carotid sinus nerves has little or no effect on the reflex response to ADN stimulation in anesthetized SD rats (12).

The brain is particularly susceptible to environmental stimuli. On the one hand, intermittent hypoxic stress in the central nervous system triggers important adaptive responses promoting neuronal survival and enabling the organism to cope with such changes. As reported, CIH induces a long-lasting augmentation of phrenic inspiratory responses in carotid-dener- vated rats (2). On the other hand, CIH leads to significant neurobehavioral deficits and cortical and hippocampal changes (2, 18, 19, 21, 22, 29, 33, 43, 50, 51). CIH-induced apoptosis in the brain is reported to be associated with oxidative stress (59).

The present findings indicate that in contrast to chemoreceptor reflex whereby the central nervous system is adjusted to adapt to hypoxic challenges, CIH induces a functional deficit to the central nervous system that may apparently contribute to the reduced baroreflex control of HR. It is likely that CIH may induce pathophysiological events (such as apoptosis) within the central autonomic nervous system leading to CIH-mediated baroreflex deficits. This assumption is supported by our recent findings (60, 61) that HR responses to microinjection of l-glutamate, NMDA, and α-amino-3-hydroxy-5-methylisox- azole-4-propionic acid (AMPA) into the left NA are all significantly reduced, and that such reduced responses are associated with changes in glutamate receptor expression (NMDA and AMPA) in the NA region of F344 rats following CIH. To-
together, these findings further support the possibility that the central autonomic nervous system is responsible for the CIH-induced impairment of baroreflex control of HR.

It should be pointed out that sleep apnea induces basal sympathetic overactivity that in turn elicits hypertension (39, 40). In the rat model, it is reported that CIH induces sympathetic overactivity as well as elevated plasma corticosterone, which may contribute to hypertension (14, 63, 64). Whether the baroreflex control of the sympathetic nerve activity is altered after CIH in the adult, and how the different components of the sympathetic system are changed to alter baroreflex of sympathetic nerve activity and hence the baroreflex control of blood pressure, are interesting issues and should be further explored. Thus far, we have only studied impaired control of renal sympathetic nerve activity following neonatal intermittent hypoxia in rats (55). It is very likely that the changes of baroreflex control of sympathetic nerve activity induced by CIH in adulthood would be different from the impairment following neonatal intermittent hypoxia. In the present study, however, we focused our experiments on the baroreceptor afferent, central, and parasympathetic efferent components within the baroreflex arc.

Summary

Our results demonstrate that CIH significantly attenuates the central mediation of baroreflex control of HR but augments baroreceptor afferent sensitivity and HR responses to vagal efferent stimulation. The enhancement of the peripheral autonomic components may represent attempts to partially compensate for the central baroreflex impairment. Since the central components in the cardiac vagal baroreflex loop include central terminals of baroreceptor afferents, neurons in NTS, and vagal motoneurons, we suggest that alterations in some or all of these components must have occurred after exposure to CIH. Therefore, the present experiments provide a foundation for more focused exploration of CIH-induced changes in barosensitive neurons in the central baroreflex circuitry.

GRANTS

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