Hypertension alters GABA receptor-mediated inhibition of neurons in the nucleus of the solitary tract

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Mei, Lin, Jing Zhang, and Steve Mifflin. Hypertension alters GABA receptor-mediated inhibition of neurons in the nucleus of the solitary tract. Am J Physiol Regul Integr Comp Physiol 285: R1276–R1286, 2003; 10.1152/ajpregu.00255.2003.—Previous studies have demonstrated that microinjection of baclofen, a GABAB receptor agonist, into the nucleus of the solitary tract (NTS) results in an enhanced pressor response in hypertensive (HT) rats compared with normotensive (NT) rats, suggesting a possible alteration in the responses of neurons in this area to activation of GABAB receptors. The following studies were designed to determine whether HT alters the sensitivity of neurons in the NTS to GABA receptor agonists. Sham-operated NT and unilateral nephrectomized, renal-wrap Sprague-Dawley rats were anesthetized, and the responses of NTS neurons receiving aortic nerve (AN) afferent inputs to iontophoretic application of GABA, the GABA\textsubscript{A} receptor agonist muscimol, and the GABA\textsubscript{B} agonist baclofen were examined. The AN input was classified as monosynaptic (MSN) if the cell responded to each of two stimuli separated by 5 ms with an action potential. If the cell did not respond, the input was considered polysynaptic (PSN). In MSNs, inhibition of AN-evoked discharge by GABA was not altered in 1 wk of HT but was reduced in 4 wk of HT, whereas in PSNs, sensitivity to GABA was reduced at 1 and 4 wk of HT. In HT rats, inhibition of AN-evoked discharge by baclofen was enhanced in MSNs, but not in PSNs, after 1 and 4 wk of HT, whereas inhibition by muscimol was reduced in MSNs and PSNs at 1 and 4 wk of HT. Changes in sensitivity to muscimol and baclofen within MSNs were the same whether the AN received a slowly or a rapidly conducted AN afferent input. The results demonstrate that early in HT the sensitivity of NTS neurons to inhibitory amino acids is altered and that these changes are maintained for \( \approx 4 \) wk. The alterations are dependent on the subtype of GABA receptor being activated and whether the neuron receives a mono- or polysynaptic baroreceptor afferent input.

the importance of GABA within the nucleus of the solitary tract (NTS) as an inhibitory modulator of the arterial baroreceptor reflex has been well established. Microinjection studies of GABA agonists and antagonists have demonstrated tonic GABAergic modulation of baroreflex function, and single-unit recording studies have demonstrated that inhibition of NTS neurons results from application of GABA\textsubscript{A} or GABA\textsubscript{B} receptor agonists (for review see Ref. 10). GABA\textsubscript{A} receptors are ligand-gated chloride ionophores that produce postsynaptic inhibition (12), and GABA\textsubscript{B} receptors are metabotropic, G protein-coupled receptors that mediate presynaptic and postsynaptic inhibition by reductions in calcium conductance or increases in potassium conductance, respectively (8). Activation of GABA\textsubscript{B} receptors on NTS neurons results in chloride-dependent membrane hyperpolarization and inhibition of baroreceptor-evoked discharge (10, 19). Activation of GABA\textsubscript{B} receptors can evoke presynaptic and/or postsynaptic inhibition of NTS neurons (1, 19) and inhibit baroreceptor-evoked discharge in NTS neurons (19).

The sensitivity of NTS neurons to the selective GABA\textsubscript{A} agonist muscimol was the same whether the neuron received a monosynaptic or a polysynaptic aortic nerve (AN) input (19). In contrast, NTS neurons receiving a monosynaptic AN input were much less sensitive to the inhibitory effects of the GABA\textsubscript{B} agonist baclofen than were NTS neurons receiving a polysynaptic AN input (19). An in vitro study reported that presynaptic inhibition of monosynaptic tractus inputs to NTS neurons occurred at a dose of baclofen approximately an order of magnitude lower than the dose necessary to induce postsynaptic inhibition (1). In most NTS neurons, stimulation of afferent inputs evokes excitation followed by postsynaptic inhibition (10). The postsynaptic inhibition is largely, but not totally, abolished by blockade of GABA\textsubscript{A} receptors (18). These results suggest that, under normal conditions, postsynaptic inhibition within the NTS is mediated primarily by activation of GABA\textsubscript{A} receptors.

Within the past decade, studies have indicated that alterations in GABAergic function within the NTS accompany chronic hypertension (HT). In several models of HT (e.g., spontaneously hypertensive rats, deoxycorticosterone acetate salt, and renal wrap), an enhanced pressor response to the microinjection of baclofen into the NTS has been observed, whereas responses to GABA\textsubscript{A} agonists remain the same as in normotensive (NT) rats (3, 15, 17). In the renal-wrap model of HT, the enhanced pressor response to NTS microinjections of the GABA\textsubscript{B} receptor agonist baclofen is associated with increased sensitivity of NTS neurons to GABA\textsubscript{B} receptor activation.

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with increased expression of GABA<sub>B</sub> receptor mRNA (3). Alterations in the blood pressure responses to GABAergic agonists suggest, but do not demonstrate, alterations in neuronal sensitivity. Changes in the sensitivity of NTS neurons to modulators such as GABA could alter baroreflex function in HT. The present study was designed to examine the sensitivity of NTS neurons receiving arterial baroreceptor afferent inputs to GABA<sub>A</sub> and GABA<sub>B</sub> receptor agonists in HT rats.

**METHODS**

**General.** Experiments were performed on male Sprague-Dawley rats (350–500 g; Charles River Laboratories, Wilmington, MA; Harlan Sprague Dawley, Indianapolis, IN). Rats were housed two per cage in a fully accredited (American Association for Accreditation of Laboratory Animal Care and US Department of Agriculture) laboratory animal room with free access to food and water. All rats were given ≥1 wk to acclimate before use. All experimental protocols were approved by the Institutional Animal Care and Use Committee and are in accordance with US Public Health Service guidelines.

**HT model.** HT was induced using a one-kidney renal-wrap procedure. Rats were anesthetized with medetomidine (0.5 mg/kg ip; Pfizer) and ketamine (75 mg/kg ip; Fort Dodge Laboratory). A Grollman renal wrap (4) and contralateral nephrectomy were performed on these animals under aseptic conditions. Control animals consisted of sham-operated rats, which were similarly anesthetized and received a unilateral nephrectomy but no wrap of the contralateral kidney, or rats that were subjected to no surgical procedures before the day of the experiment. Because the responses of both control groups of rats were identical, they were grouped together for analysis. Anesthesia was terminated by atipamezole (1 mg/kg ip; Pfizer) at the conclusion of the surgical procedures. Postoperative analgesics [nalidixic acid (Nubaine), administered intramuscularly] were available as needed.

**Acute surgical preparation.** Rats were initially anesthetized with pentobarbital sodium (60 mg/kg ip) and placed on a thermostatically controlled heating pad. Body temperature was maintained at 36–38°C throughout the experiment. After placement of a venous catheter (tail vein) and cannulation of the trachea, the animal was artificially ventilated with oxygenated room air, and subsequent anesthetic was given as an infusion of 10–20 mg·kg<sup>−1</sup>·h<sup>−1</sup> iv. Gallamine triethiodide (20 mg·kg<sup>−1</sup>·h<sup>−1</sup> iv) was given for paralysis to reduce respiratory movements of the brain stem. A femoral artery was cannulated for arterial blood pressure monitoring (CDX transducer, Cobe Laboratories, Lakewood, CO). Mean arterial pressure and heart rate were determined from the pulsatile signal using a Coulburn blood pressure processor. Depth of anesthesia was assessed by monitoring the stability of arterial pressure and heart rate in response to pinch of the hindpaw and was adjusted by appropriate changes in the infusion rate. The ANs were isolated bilaterally and marked with small pieces of black suture. The rat was placed in a stereotaxic head frame, and an occipital craniotomy was performed to expose the dorsal surface of the medulla in the region of the calamus scriptorius. The AN ipsilateral to the central recording site was mounted on bipolar stimulating electrodes and covered with a mixture of petroleum jelly and mineral oil.

**Extracellular recordings and iontophoresis.** Extracellular action potential discharge was recorded with a five-barrel electrode. The tip of the electrode was broken to give an outer diameter of ≤1 μm. The recording barrel was filled with a solution of 0.5 M sodium acetate containing 2% Chicago sky blue (impedance 10–40 MΩ). One barrel of each five-barrel electrode was filled with a solution of 3 M NaCl and was used for automatic current balancing, control injections of unbalanced current, and pH control measurements. The remaining barrels were filled with different drug solutions. All recordings were performed within a region 1.2 mm caudal and 1.0 mm rostral to the calamus scriptorius, 0–0.8 mm lateral to the midline, and 0.2–1 mm below the surface. The electrode was lowered into the tissue in 2.0- to 2.5-μm steps by a stepper driver (Burleigh Instrument, Fishers, NY). The AN was stimulated with single pulses (1-ms duration, 0.5 Hz, 500 μA). Recorded action potentials were amplified by a direct-current amplifier (World Precision Instrument, New Haven, CT), passed through an alternating-current filter, and then sent to a digital oscilloscope (Nicolet Instrument, Madison, WI), an audio monitor (Grass Instrument, Quincy, MA), a videotape recorder (Vetter, Reberburg, PA), and a window discriminator (World Precision Instrument, Sarasota, FL). The window discriminator output was led to an analog-to-digital converter (model CED1401, Cambridge Electronics, Cambridge, UK) interfaced with a personal computer. Spike2 data acquisition software (Cambridge Electronic Design) was used for on- and off-line analyses. After an AN-evoked NTS neuron was found, monosynaptic neurons (MSNs) or polysynaptic neurons (PSNs) were characterized using the ability of the AN-evoked responses of MSNs to follow two stimuli separated by 5 ms (13). Peristimulus-time histograms (PSTHs; 1-ms bin width, 40 sweeps of AN stimulation at 0.5 Hz) and rate meter histograms (180-s duration, 1-s bin width) were collected to analyze evoked and spontaneous discharge, respectively.

**After baseline discharge (spontaneous firing rate and/or evoked discharge) was recorded, drugs were administered by application of microiontophoretic ejecting currents to the drug-containing barrels. For current-response curves, drugs were ejected for successive 10-s periods separated by 10- to 20-s intervals. Responses were measured under steady-state conditions. Typically, three to four ejection cycles were required to reach a steady state. The drug solutions for microiontophoresis were GABA (2 mM, pH 4.5; Research Biochemicals International, Natick, MA), muscimol (0.04 mM, pH 4.5; RBI), and baclofen (10 mM, pH 4.5; RBI). All drugs were dissolved in 150 mM NaCl, and were ejected as cations. Retaining currents of appropriate polarity were applied to the drug barrels to reduce the passive diffusion of the drug from the electrode tip during nonejection periods. All currents ejected by ejecting barrels were automatically balanced by the iontophoretic unit (Neurophone, Medical Systems) by ejection of opposite currents through the balancing barrel. After a successful recording, dye was iontophoretically ejected from the recording barrel, and standard postmortem histological procedures were performed to localize the recording site.

**Data analysis.** Iontophoretic “dose-response” curves were constructed from PSTHs of responses to 40 AN stimuli obtained before, during, and 2–5 min after iontophoretic application of agonist at ejection currents of 5, 10, 20, and 40 nA, as previously described (19) (Fig. 1). Data were accepted only if AN-evoked discharge in the recovery PSTH was within ±10% of the discharge in the control PSTH obtained before iontophoresis of agonist. It was not possible to test every drug on each cell. However, for each drug, all ejection currents were tested on each cell presented in the dose-response curves. To account for different levels of basal spontaneous...
discharge, baseline discharge was measured in the absence of AN stimulation, and this value was subtracted from AN-evoked discharge. Response onset latency was defined as the interval between the stimulus artifact and the bin containing the greatest number of action potentials. Differences in AN-evoked discharge at each iontophoretic ejection current were tested between NT, 1-wk-HT, and 4-wk-HT rats using ANOVA with Student-Newman-Keuls method for post hoc comparisons. For statistical analysis, the absolute number of AN-evoked or spontaneous action potentials was used; for graphical purposes, these values were normalized to the 100% control level of discharge observed in the absence of any drug application. Values are means ± SE, and significance was accepted at \( P < 0.05 \). All values of \( n \) refer to the number of neurons, except where mean arterial pressure data are reported, in which case \( n \) refers to the number of rats.

**RESULTS**

The mean arterial pressure under resting conditions for the NT rats was 107 ± 5 mmHg (\( n = 45 \)). In NT rats, data were obtained from 105 AN-evoked NTS neurons: 42 were characterized as receiving monosynaptic inputs from AN afferents (MSNs), and 63 were characterized as receiving polysynaptic AN inputs (PSNs). The mean arterial pressure under resting conditions for the HT rats was 151 ± 4 mmHg after 1 wk (\( n = 29 \)) and 149 ± 5 mmHg after 4 wk (\( n = 36 \)). In the 1-wk-HT rats, data were obtained from 49 AN-evoked NTS neurons: 19 were characterized as MSNs and 30 as PSNs. In the 4-wk-HT rats, data were obtained from 65 AN-evoked NTS neurons: 25 were characterized as MSNs and 40 as PSNs.

Spontaneous action potential discharge was not observed in all neurons. In those neurons that did discharge spontaneously, there was no difference in discharge rate (action potentials per second) within each MSN and PSN group among neurons from NT [4.4 ± 0.5 (\( n = 29 \)) for MSN and 6.7 ± 0.7 (\( n = 45 \)) for PSN], 1-wk-HT [4.9 ± 0.6 (\( n = 14 \)) for MSN and 6.5 ± 0.9 (\( n = 24 \)) for PSN], and 4-wk-HT [4.1 ± 0.5 (\( n = 19 \)) for MSN and 7.1 ± 0.9 (\( n = 30 \)) for PSN] rats. MSNs had a lower spontaneous discharge frequency than PSNs in NT (\( P = 0.02 \)) and 4-wk-HT (\( P = 0.02 \)) rats.

Under control conditions, the number of action potentials evoked by 40 AN stimuli was not different...
between MSN and PSN groups in NT \( [34 \pm 2 (n = 42) \) for MSN and \( 32 \pm 1 (n = 63) \) for PSN], 1-wk-HT \( [30 \pm 1 \) for MSN \( (n = 19) \) and \( 29 \pm 1 \) for PSN \( (n = 30) \), and 4-wk-HT \( [37 \pm 3 \) for MSN \( (n = 25) \) and \( 34 \pm 2 \) for PSN \( (n = 40) \) rats. Within each MSN and PSN group, there was no difference between the number of action potentials evoked by 40 AN stimuli in neurons from NT rats and the number evoked in neurons from 1- or 4-wk-HT rats.

Figure 2 illustrates the onset latency distribution of MSNs and PSNs from the NT and HT rats. The latencies for neurons recorded in 1- and 4-wk-HT rats were combined. The mean onset latency for the entire population of MSNs was \( 16.8 \pm 1.5 \) ms in NT rats \( (n = 42) \) and \( 19.1 \pm 1.6 \) ms in HT rats \( (n = 44) \). However, the MSN population in NT and HT rats was clearly bimodal. The onset latency of approximately one-third of the MSNs fell between 2 and 8 ms \( [4.3 \pm 0.6 \) ms \( (n = 16) \) in NT and \( 4.7 \pm 0.5 \) ms \( (n = 14) \) in HT, \( P > 0.05 \). Conduction distance was estimated at 5–6 cm, suggesting that these MSNs received AN afferent inputs conducting at \(-10–30 \) m/s. The onset latency for the remaining MSNs was 16–36 ms \( [23.7 \pm 0.7 \) ms \( (n = 26) \) in NT and \( 25.9 \pm 0.9 \) ms \( (n = 30) \) in HT, \( P > 0.05 \), suggesting that these cells received afferent inputs conducting at 1–4 m/s. The latency distribution for PSNs was unimodal, and there was no difference between the latencies of the NT \( (26.1 \pm 1.2 \) ms, \( n = 63) \) and HT \( (27.4 \pm 1.2 \) ms, \( n = 70) \) PSNs \( (P > 0.05) \). Reconstruction of the recording sites indicated no diff-

Fig. 2. Onset latency distributions in neurons receiving monosynaptic AN inputs (MSNs) and polysynaptic AN inputs (PSNs) in normotensive (NT) and hypertensive (HT) rats. Numbers in parentheses refer to number of neurons in each group.
Sensitivity to GABA. In the MSN population, there was no difference in the inhibition of AN-evoked discharge by GABA between 1-wk-HT (n = 8) and NT (n = 13) rats at any of the ejection currents tested (Fig. 3). In 4-wk-HT rats (n = 8), the inhibition of AN-evoked discharge in MSNs was significantly attenuated at all ejection currents compared with NT and 1-wk-HT rats. In the PSN population the inhibition by GABA was reduced in 1-wk-HT rats at ejection currents of 20 and 40 nA (n = 8), and in 4-wk-HT rats the inhibition of AN-evoked discharge was less at all ejection currents (n = 17) than in NT rats (n = 17; Fig. 3). In NT rats, AN-evoked discharge in MSNs was inhibited less than in PSNs during GABA iontophoresis at 20 and 40 nA (P < 0.05). This difference was not observed in 1- and 4-wk-HT rats.

Sensitivity to muscimol. In the MSN population, the inhibition of AN-evoked discharge by muscimol was less in 1-wk-HT (n = 8) and 4-wk-HT (n = 9) than in NT (n = 12) rats at all ejection currents (Fig. 4). A similar observation was made in the PSN population; however, the attenuation of muscimol inhibition in 1-wk-HT rats was not significant at the 5-nA ejection current (n = 20, 6, and 9 in NT, 1-wk HT, and 4-wk-HT rats, respectively). In the MSNs and PSNs, muscimol inhibition of AN-evoked discharge in neurons from
1-wk-HT rats was not different from that in neurons from 4-wk-HT rats. In NT and HT rats, there was no difference in the inhibition of AN-evoked discharge between MSNs and PSNs.

**Sensitivity to baclofen.** In the MSN population, the inhibition of AN-evoked discharge by baclofen at all ejection currents was greater in 1-wk-HT \((n = 6)\) and 4-wk-HT \((n = 6)\) than in NT \((n = 13)\) rats (Fig. 5). In the PSN population, the sensitivity to baclofen was the same in 1-wk-HT \((n = 7)\) and 4-wk-HT \((n = 12)\) rats compared with NT rats \((n = 23)\). In the MSNs and PSNs, baclofen inhibition of AN-evoked discharge in neurons from 1-wk-HT rats was not different from the inhibition in neurons from 4-wk-HT rats. In NT rats, baclofen iontophoresis inhibited AN-evoked discharge less in MSNs than in PSNs at all ejection currents \((P < 0.05)\). This difference was abolished in 1-wk-HT rats and reversed in 4-wk-HT rats, so that baclofen inhibited MSNs more than PSNs.

**MSNs receiving slowly vs. rapidly conducted AN inputs.** As illustrated in Fig. 2, the distribution of onset latencies for MSNs was bimodal. Sensitivity to muscimol and baclofen was examined in MSNs receiving slowly vs. rapidly conducting AN afferent inputs. Data from 1- and 4-wk-HT rats were combined. The reduced inhibition by muscimol and the enhanced inhibition by baclofen was observed in MSNs, regardless of the apparent conduction velocity of the afferent input: in NT rats receiving rapidly conducting inputs, \(n = 5\) for muscimol and baclofen; for NT rats receiving slowly conducting inputs, \(n = 7\) for muscimol and \(n = 8\) baclofen; in HT rats receiving rapidly conducting inputs, \(n = 8\) for muscimol and \(n = 6\) for baclofen; in HT rats receiving slowly conducting inputs, \(n = 10\) for muscimol and \(n = 12\) baclofen.
conducting inputs, \( n = 9 \) for muscimol and \( n = 6 \) for baclofen (Fig. 6).

**Responses of nonevoked neurons.** During the course of the examination of NTS neurons receiving AN inputs, a number of recordings were obtained from spontaneously discharging neurons that did not respond to the AN stimulus. The inhibition of the spontaneous discharge of these non-AN-evoked neurons was examined in NT, 1-wk-HT, and 4-wk-HT rats. There was no difference in GABA inhibition in non-AN-evoked neurons recorded between HT (\( n = 8 \) and 40 for 1- and 4-wk-HT rats, respectively) and NT (\( n = 26 \)) rats (Fig. 7). There was a trend toward reduced muscimol inhibition in non-AN-evoked neurons recorded in HT rats (\( n = 14 \) and 25 for 1- and 4-wk-HT rats, respectively); however, it did not reach statistical significance compared with non-AN-evoked neurons recorded in NT rats (\( n = 43 \)). Baclofen inhibition was enhanced in non-AN-evoked neurons recorded in 1-wk-HT rats (\( n = 8 \)) at 40-nA ejection current and in neurons recorded in 4-wk-HT rats (\( n = 41 \)) at 20 and 40 nA compared with NT rats (\( n = 56 \)).

**DISCUSSION**

**General.** The results demonstrate that the inhibition of NTS neurons receiving arterial baroreceptor inputs by GABA receptor agonists is altered in HT. These alterations occur within a short, 1-wk, period and persist for \( \geq 4 \) wk after induction of HT. Previous studies have demonstrated enhanced responses to NTS microinjection of the GABA\(_B\) receptor
agonist baclofen in this (3, 17) and other (15) models of HT. This finding is consistent with the enhanced inhibition of monosynaptic AN-evoked discharge in NTS neurons by iontophoretic application of baclofen. In NT rats, baclofen inhibited AN-evoked discharge less in MSNs than in PSNs, as we previously reported (19). Within 1 wk of HT, MSN sensitivity to baclofen is comparable to that in the PSNs, and by 4 wk, MSNs are more sensitive than PSNs. These changes occur regardless of the conduction velocity of the afferent input.

MSNs exhibit reduced muscimol inhibition and enhanced baclofen inhibition after 1 wk of HT; therefore it is not clear why GABA inhibition of MSNs was not altered after 1 wk of HT. This could reflect the relative proportion of GABA\(_A\) to GABA\(_B\) receptors on a neuron after 1 wk of HT, relative binding affinities, differences in desensitization produced by GABA compared with muscimol, or any number of other factors that cannot be differentiated in the present study.

In contrast to GABA\(_B\) receptor-mediated inhibition, previous studies have not found alterations in the responses to microinjection of muscimol in HT rats (3, 15). Therefore, one might predict that the sensitivity of AN-evoked neurons to muscimol would not be altered in HT rats. However, we found that MSNs and PSNs are less sensitive to muscimol-evoked inhibition within 1 wk of HT. The attenuated inhibition is present and quantitatively similar after 4 wk of HT. Microinjections will influence the total population of neurons that ultimately subserve a sympathoinhibitory function in a given area: some that receive AN inputs and some that do not. The data presented in Fig. 7 suggest that muscimol-evoked inhibition of non-AN-evoked neurons, some of which may subserve sympathoinhibitory functions, is not altered in HT rats. Normal responses
Fig. 7. GABA (A), muscimol (B), and baclofen (C) inhibition of spontaneous discharge in neurons that did not respond to AN stimulation in NT, 1-wk-HT, and 4-wk-HT rats. Numbers in parentheses refer to number of neurons in each group.
to microinjections of muscimol within the non-AN-evoked neuron population could mask reduced inhibition within the population of AN-evoked neurons. The method for presumptive identification of NTS neurons receiving monosynaptic inputs used in this study (13) has previously identified MSNs receiving rapidly or slowly conducting AN afferent inputs (19, 21). The present report reinforces and extends the previous observation that sensitivity to muscimol or baclofen is similar, regardless of the conduction velocity of the afferent input (19), by demonstrating that the alterations in sensitivity to GABA agonists are similar, regardless of the conduction velocity of the afferent input.

The sensitivity of non-AN-evoked NTS neurons to GABA<sub>A</sub> receptor activation was not altered; however, at higher ejection currents, sensitivity of these neurons to GABA<sub>B</sub> receptor activation was enhanced, as in the AN-evoked neurons, albeit to a lesser degree. It is quite likely that some percentage of the non-AN-evoked neurons consists of neurons that receive baroreceptor inputs from the carotid sinus and/or cardiopulmonary vagal afferent inputs, and these neurons might also exhibit altered sensitivity to GABAergic agonists in HT, as do neurons receiving AN baroreceptor afferent inputs. Depending on the percentage of the total population that these non-AN-evoked neurons receiving other baroreceptor afferent inputs comprise, they could influence the population response. Alternatively, the alterations in sensitivity to GABA<sub>B</sub> agonist described in this report might represent a more general change within the NTS whether or not the neuron receives a baroreceptor input.

Mechanism of alterations. It is appropriate to speculate on the possible triggers that initiate the alterations in sensitivity to GABAergic agonists observed in this study. Enhanced excitatory (7) or inhibitory synaptic input and/or circulating hormones (e.g., angiotensin and catecholamines) (5, 6, 16) could serve to initiate the alterations. Modulation of GABA<sub>A</sub> receptor function can be the result of any number of potential mechanisms, including, but not restricted to, regulation of receptor number and/or affinity, regulation of the phosphorylation status of the receptor, and alterations in receptor subunit composition (12). For example, prolonged exposure to GABA downregulates the GABA<sub>A</sub> receptor (12). In addition, GABA<sub>A</sub> receptor currents are suppressed during concomitant application of N-methyl-D-aspartate (2) or brief (1-s) depolarizations of the postsynaptic membrane (11). These results suggest that increased synaptic input, either excitatory or inhibitory, could play a role in the reduced sensitivity to muscimol.

In comparison to GABA<sub>A</sub> receptors, little is known about potential mechanisms that may alter GABA<sub>B</sub> receptor function. Prolonged exposure to GABA induces expression of GABA<sub>B</sub> receptors in cerebellar granule cells (14). Increased nociceptive afferent inputs from the hindlimb increased the expression of the GABA<sub>B</sub> mRNA in dorsal root ganglia and dorsal regions of the lumbar spinal cord (9), suggesting that synaptic activity can regulate the expression of GABA<sub>B</sub> receptors. We have reported that HT is associated with an increased expression of GABA<sub>B</sub> receptor mRNA (3) and provided indirect evidence that this change occurs primarily in postsynaptic, as opposed to presynaptic, elements of the NTS (17). This suggests an enhanced expression of GABA<sub>B</sub> receptors, and our data further suggest that this expression occurs in MSNs.

Physiological significance of alterations. The results demonstrate that within 1 wk of HT there is a shift in the balance of sensitivity to receptor-selective GABAergic agonists and that these alterations persist for ≥4 wk after the induction of HT. In the NT rat, AN-evoked excitatory inputs are often followed by inhibitory inputs, and this inhibition is mediated predominantly by GABA<sub>A</sub> receptors (10), with GABA<sub>B</sub> receptors playing a relatively minor role (18). In previous work (19) and the present study, this is mirrored in the sensitivity of MSNs, inasmuch as they are very sensitive to muscimol inhibition but not very sensitive to baclofen. In MSNs in the HT rat, this GABAergic sensitivity profile is reversed.

GABA<sub>A</sub>-mediated inhibition through ionotropic chloride channels is relatively rapid in onset and short in duration, whereas GABA<sub>B</sub> inhibition through G protein-coupled mechanisms is slower in onset and longer lasting. As a consequence, tonic inhibition via GABA<sub>A</sub> receptors may require greater energy expenditure or may be less effective than inhibition mediated by GABA<sub>B</sub> receptors. It has been suggested that in renal HT there is an increase in baroreceptor excitatory inputs, particularly from unmyelinated afferent fibers (7). A shift from predominantly GABA<sub>A</sub>-to GABA<sub>B</sub>-mediated inhibition may provide a more metabolically favorable mechanism to offset increased baroreceptor excitatory inputs to NTS neurons in HT rats (7). Our previous work (20) and the present study found no difference in the spontaneous discharge of AN-evoked NTS neurons between NT and HT rats; therefore, enhanced GABA<sub>B</sub> inhibition may normalize the discharge of NTS neurons receiving increased baroreceptor inputs in HT. It has been suggested that such adaptations enable the neurons to respond normally to changes in arterial pressure (10) and limit reductions in baroreflex performance in HT.

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DISCLOSURES

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REFERENCES


