Modulation of baroreceptor activity by gene transfer of nitric oxide synthase to carotid sinus adventitia

Silvana S. Meyrelles,1 Ram V. Sharma,2,4 Hui Z. Mao,2 Francois M. Abboud,2,3,5 and Mark W. Chapleau2,3,6

1Department of Physiological Sciences, Federal University of Espirito Santo, Vitoria, ES, Brazil 20042-755; 2The Cardiovascular Center and the Departments of Internal Medicine, Anatomy and Cell Biology, and 3Physiology and Biophysics, The University of Iowa, Iowa City 52242; and the 4Veterans Affairs Medical Center, Iowa City, Iowa 52246

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Modulation of baroreceptor activity by gene transfer of nitric oxide synthase to carotid sinus adventitia. Am J Physiol Regul Integr Comp Physiol 284: R1190–R1198, 2003; 10.1152/ajpregu.00735.2002.—Administration of nitric oxide (NO) or NO donors to isolated carotid sinus and carotid bodies inhibits the activity of baroreceptor and chemoreceptor afferent nerves. Furthermore, NO synthase (NOS) is present in endothelial cells and in sensory nerves innervating the carotid sinus region. The major goal of this study was to determine whether overexpression of NOS in carotid sinus modulates baroreceptor activity. Rabbits were anesthetized, and adenoviral vectors (5 × 108 plaque-forming units) encoding genes for either β-galactosidase (β-Gal) or endothelial type III NOS (eNOS) were applied topically to the adventitial surface of one carotid sinus. In some experiments, the NOS inhibitor N(ω)-nitro-arginine methyl ester (L-NAME) was applied to the carotid sinus immediately after the vector. Four to five days later, baroreceptor activity and carotid sinus diameter were measured from the vascularly isolated carotid sinus of the anesthetized rabbits. Transgene expression was confirmed by X-Gal staining of β-Gal and measurement of NOS activity by citrulline assay. The expression was restricted to the carotid sinus adventitia. Baroreceptor activity was decreased significantly, and the pressure-activity curve was shifted to higher pressures in eNOS-transduced (n = 5) compared with β-Gal-transduced (n = 5) carotid sinuses. The pressure corresponding to 50% of maximum activity averaged 55 ± 6 and 76 ± 7 mmHg in β-Gal- and eNOS-transduced carotid sinuses, respectively (P < 0.05). Decreased baroreceptor activity was accompanied by a significant increase in carotid diameter in the eNOS-transduced carotid sinuses (n = 5). L-NAME prevented the inhibition of baroreceptor activity and the increase in carotid diameter in eNOS-transduced carotid sinuses (n = 5). We conclude that adenoviral-mediated gene transfer of eNOS to carotid sinus adventitia causes sustained, NO-dependent inhibition of baroreceptor activity and resetting of the baroreceptor function curve to higher pressures.

NITRIC OXIDE (NO) is an important autocrine/paracrine factor that mediates vascular relaxation, signaling within the nervous system, and immunological responses (28). Type III endothelial NO synthase (eNOS) and type I neuronal NOS (nNOS) isoforms are present in vascular endothelial cells and sensory nerves innervating the carotid sinus region, respectively (9, 14, 41). Furthermore, NO inhibits chemoreceptor and baroreceptor afferent nerve activity acutely (25, 33, 38). The functional role of endogenous NO and its potential to chronically modulate baroreceptor sensitivity are unknown.

The major goal of the present study was to determine whether sustained overexpression of NOS in the carotid sinus modulates carotid sinus baroreceptor activity. Localized overexpression of NOS was accomplished by adenoviral-mediated gene transfer of eNOS to the carotid sinus (5, 26, 34).

MATERIALS AND METHODS

A total of 32 New Zealand White rabbits of either sex were studied. Experimental procedures were carried out in accordance with the American Physiological Society’s “Guiding Principles for Research Involving Animals and Human Beings” and institutional guidelines. One carotid sinus from each rabbit was subjected to gene transfer of either the reporter gene β-galactosidase (β-Gal) or bovine type III eNOS. In some experiments the NOS inhibitor N(ω)-nitro-arginine methyl ester (L-NAME) was applied to the carotid sinus immediately after application of the viral vector suspension. The contralateral carotid artery and carotid sinus region (nontransfected) provided additional controls for some of the protocols.

Adenoviral Vectors

Adenoviral vectors were constructed in the University of Iowa Gene Transfer Vector Core Laboratory (7, 8, 12, 26, 31, 34). The cDNAs for the reporter gene β-galactosidase (β-Gal) and type III eNOS were cloned into shuttle vectors containing sequences from serotype 5 human adenovirus, and the Rous sarcoma virus (RSV) promoter was used to drive transcription. The transgene plasmid was transfected into human embryonic kidney 293 cells along with a plasmid containing the serotype 5 human adenovirus genome with dele-
Topical Application of Vectors to Carotid Sinus

Rabbits were anesthetized with intramuscular injection of xylazine (20 mg/kg) and ketamine (55 mg/kg). Using sterile surgical technique, one carotid sinus was exposed via a small incision in the cervical region. Approximately 50 μl of adenoviral vector suspension (5 × 10⁸ plaque-forming units (pfu/ml), stored at −70°C.

Analysis of Transgene Expression

β-Gal transgene expression was confirmed 4–5 days after application of the β-Gal vector to the carotid sinus by histological analysis (8, 26, 34). The carotid sinus region was removed, rinsed with phosphate-buffered saline, and fixed in glutaraldehyde (0.5%) for 30 min at room temperature. The carotid sinuses were then washed with phosphate-buffered saline containing MgCl₂ (1 mM) and exposed to K₂Fe(CN)₆ (4.9 mM), K₃Fe(CN)₆ (4.7 mM), and 5-bromo-4-chloro-3-indolyl-β-d-galactopyranoside (X-Gal, 2.4 mM) in saline containing MgCl₂ for 1 h at 37°C. The X-Gal solution was then replaced with saline, and the blood vessels were stored at 4°C. The same procedure was performed on the contralateral, nontransfected carotid sinuses.

The citrulline assay (3, 8, 12) was used to quantify NOS activity in four NOS-transduced and four contralateral control carotid sinuses removed from four rabbits 4–5 days after application of the NOS vector to one carotid sinus. The carotid sinuses were homogenized on ice using a glass homogenizer fitted with a ground glass pestle in 50 mM Tris·HCl (pH 7.4) containing 0.1 mM EDTA, 0.1 mM EGTA, 12 mM 2-mercaptoethanol, 1 mM phenylmethylsulfonyl fluoride, 3 μM leupeptin, 1 μM aprotinin, and 1 μM pepstatin, and 1 μM soybean trypsin inhibitor. The assay mixture contained 50 mM Tris·HCl (pH 7.4), 5 μM l-arginine, 0.25 μCi l-[³H]arginine, 0.5 mM α-NADPH, 10 μM tetrahydrobipterin, 4 μM flavin mononucleotide, 4 μM FAD, 1 μg calmodulin, 1 mM calcium, and 40–80 μg cell homogenate protein in 200 μl volume. To determine calcium-independent NOS activity, calcium was replaced by 1 mM EGTA. To confirm that the intensity measured reflected NOS activity, the NOS inhibitor N⁵-nitro-l-arginine (l-NNA, 1 mM) was added to the assay. Enzyme assays were carried out at 37°C for 15 min and terminated by adding 5.5 ml of Dowex slurry (Dowex AG50W-X8, 100- to 200-mesh sodium form). l-[³H]citrulline production was measured by using a liquid scintillation spectrometer (Beckman-Coulter, model DU 640, Brea, CA).

Measurement and Analysis of Baroreceptor Pressure-Activity Relation

Baroreceptor activity was measured from isolated carotid sinuses subjected to β-Gal gene transfer (n = 5), NOS gene transfer (n = 5), NOS gene transfer plus l-NNAME treatment (n = 5), and β-Gal gene transfer plus l-NNAME treatment (n = 5). Four to five days after topical application of vector to the carotid sinus, rabbits were anesthetized with pentobarbital sodium (30–35 mg/kg iv) and mechanically ventilated with room air supplemented with oxygen. Ventilation was adjusted to maintain arterial blood gases and pH within normal values. Polyethylene catheters were placed into the femoral artery and vein for measurement of arterial pressure and administration of anesthetic, respectively.

The carotid sinus subjected to gene transfer 4–5 days beforehand was vascularily isolated as described previously (24–26). Briefly, the common, external, and internal carotid arteries were isolated and ligated along with other arterial branches, and catheters were placed in the common carotid and lingual arteries, thereby isolating the carotid sinus lumens from the adjacent circulation. The isolated carotid sinus was filled with an oxygenated Krebs-Henseleit buffer. Carotid sinus pressure was measured with a transducer (model P231D, Statham) connected to the lingual artery catheter and was varied in a controlled manner by altering the inflow of air from a pressurized air source into a glass fluid reservoir attached to the common carotid artery catheter. The cervical sympathetic, aortic depressor, and vagus nerves were sectioned on the side of the isolated carotid sinus. Skeletal muscle contractions were prevented during periods of nerve recording by administration of the neuromuscular blocker decamethonium bromide (0.3 mg/kg iv).

The carotid sinus nerve was identified, sectioned, and draped over a unipolar electrode. The electrode and nerve were insulated by covering the area with warm paraffin oil (37°C) and/or by encasing the nerve in Wackers silicone gel (24–26). Nerve activity was recorded by using a high-impedance probe (model H1151J, Grass Instrument) and a Grass band-pass amplifier (model P511J, 100- to 3000-Hz bandwidth). The nerve recording was displayed on a dual-beam storage oscilloscope (model 5113, Tektronix). A nerve traffic analyzer (model 706C, Dept. of Bioengineering, Univ. of Iowa, Iowa City, IA) was used to count the frequency of action potentials with amplitude exceeding a selected voltage level set just above the electrical noise. Systemic arterial pressure, carotid sinus pressure, and the output of the nerve traffic analyzer were continuously recorded on a chart recorder (model 11–1202–25, Gould).

Baroreceptor activity was recorded during slow ramp increases in nonpulsatile carotid sinus pressure from 0 to 160 mmHg. Pressure ramps were repeated approximately once every 5 min until baroreceptor responses were consistent. Carotid sinus pressure was held at 80 mmHg in between pressure ramps. The rate of increase in carotid sinus pressure during the ramps was similar in all experiments and averaged approximately 2.5–3.0 mmHg/s.

Baroreceptor activity was measured at 20-mmHg increments over a range of 20–160 mmHg. The data from three or four pressure ramps were analyzed, and the average responses were calculated for each experiment. Because the absolute level of multifiber baroreceptor activity is dependent on the recording conditions and varies from preparation to preparation, activity is expressed as a percentage of the maximum activity recorded at high carotid sinus pressure up to 160 mmHg. The position of the baroreceptor function curve along the pressure axis was determined directly from the experimental traces in individual experiments by measuring the carotid sinus pressure corresponding to 50% of the maximum baroreceptor activity (EP₅₀).
Measurement and Analysis of Carotid Pressure-Diameter Relation

The diameter of the carotid artery at the origin of the carotid sinus bifurcation was measured during ramp increases in carotid sinus pressure from 0 to 160 mmHg using a video micrometer (model VIA-100, Boeckeler Instruments, Tucson, AZ) (24, 25). Carotid sinuses exposed to the β-Gal vector (n = 4), the NOS vector (n = 4), and the NOS vector plus L-NAME (n = 5) were studied 4–5 days after topical application of the vectors to the carotid sinus. The carotid sinus region was viewed under magnification (×16) through a stereomicroscope (model M3C, Wild, Heerbrugg, Switzerland) during slow ramp increases in carotid sinus pressure. The image was recorded on videotape using a camera (model JE7362, Javelin Electronics, Torrance, CA), videocassette recorder (model SLV-585HF, Sony, Japan), and video monitor (model BWM12, Javelin Electronics). A digital readout of the carotid sinus pressure was simultaneously projected on the same video monitor as the carotid sinus image with the use of a beam splitter (model MPS-50, Image Labs, Pearl River, NY). The video micrometer system enabled detection of 12-μm changes in diameter over an absolute range of 0–5 mm. The compliance of the carotid sinus was estimated by calculating the slope of the pressure-diameter relation over its steepest portion between 20 and 80 mmHg using linear regression.

Carotid Sinus Histology

Both common carotid arteries and the carotid sinus regions were removed bilaterally from rabbits subjected to β-Gal or eNOS gene transfer to one carotid sinus. The unpressurized arteries were covered in OCT compound (Miles Scientific, Elkhart, IN) and frozen in a cryostat. Cross-sections (20-μm thickness) through the common carotid artery just below the bifurcation and through the carotid sinus and external carotid artery were placed on polylysine-coated slides and either processed for X-Gal staining (see Analysis of Transgene Expression) or stained with hematoxylin or eosin and photographed (Diaphot 300, Nikon, Japan).

Statistical Analysis

All data are expressed as means ± SE. Effects of carotid sinus pressure, treatment group (eNOS vs. β-Gal gene transfer and effect of L-NAME), and the interaction between pressure and treatment on baroreceptor activity and carotid diameter were analyzed by two-factor ANOVA (GB-Stat 6.0 software). When the ANOVA was significant, differences between two sets of data points were determined by Fisher’s least significant difference post hoc test (GB-Stat 6.0 software). Effects of NOS gene transfer and administration of L-NAME on baroreceptor EP50 were analyzed by unpaired t-test. Differences were considered significant when P < 0.05.

RESULTS

Transgene Expression in Carotid Sinus

Topical perivascular application of the adenoviral vectors Adβ-Gal and AdeNOS to carotid sinus led to significant expression of the transgenes in carotid sinus measured 4–5 days later. The gene transfer, visualized by X-Gal staining of β-Gal, was localized to the region of the carotid sinus and was selective to cells in the adventitia (Fig. 1). Transgene expression was not observed in the media layer of carotid artery walls or within carotid sinus nerve fibers. NOS enzymatic activity was ~100 fold higher in carotid sinuses exposed to AdeNOS (n = 4) compared with the contralateral...
control carotid sinuses \((n = 4)\) (Fig. 2). The NOS activity was essentially abolished by either replacement of calcium with the calcium chelator EGTA or addition of the NOS inhibitor L-NNA to the assay, confirming that the measured activity was indeed calcium-dependent NOS activity (Fig. 2).

**Gene Transfer and L-NAME Effects on Baroreceptor Pressure-Activity Relation**

Baroreceptor activity was inhibited significantly, and the pressure-activity curve was shifted to higher pressures in eNOS compared with β-Gal-transduced carotid sinuses (Figs. 3 and 4A). Consequently, the carotid sinus pressure corresponding to EP_{50} was significantly higher in the eNOS-transduced carotid sinuses (Figs. 3 and 4B). In a third group of rabbits, local application of the NOS inhibitor L-NAME to carotid sinus adventitia immediately after application of the AdeNOS vector completely prevented the eNOS-induced inhibition of baroreceptor activity and the increase in EP_{50} observed 4–5 days later (Fig. 4).

A possible role of endogenous NO in modulation of baroreceptor activity was investigated in a fourth group of rabbits that underwent β-Gal gene transfer and L-NAME treatment of one carotid sinus. Baroreceptor activity tended to be higher in the β-Gal-transduced sinuses treated with L-NAME \((n = 5)\) compared with untreated sinuses \((n = 5)\), but the difference did not reach statistical significance (Fig. 5). The baroreceptor function curves for L-NAME-treated β-Gal and L-NAME-treated eNOS-transduced carotid sinuses were essentially superimposable and not significantly different from each other (Fig. 5). Combining the data from these two groups enabled demonstration of a significant increase in baroreceptor activity at low levels of carotid sinus pressure (20–60 mmHg) in L-NAME-treated carotid sinuses \((n = 10)\) compared with untreated β-Gal-transduced carotid sinuses \((n = 5)\).

**Gene Transfer and L-NAME Effects on Carotid Pressure-Diameter Relation**

The carotid pressure-diameter relation was measured to evaluate possible effects of eNOS gene transfer on vascular tone and structure that might indirectly influence baroreceptor activity. The diameter of eNOS-transduced carotid sinuses was increased significantly over a wide range of pressure compared with the diameter of β-Gal-transduced sinuses (Fig. 6). Treatment of eNOS-transduced sinuses with L-NAME prevented the increase in diameter (Fig. 6). The compliance of the carotid sinus (change in diameter/change in pressure) was not significantly different in β-Gal-transduced \((14.5 ± 0.7 \mu m/mmHg, r = 0.99)\) and eNOS-transduced \((18.5 ± 2.3 \mu m/mmHg, r = 0.99)\) carotid sinuses.

![Fig. 3. Original tracings of carotid sinus pressure and baroreceptor activity recorded from a β-Gal-transduced carotid sinus (A and B) and an eNOS-transduced carotid sinus (C and D) during application of pressure ramps. The method of determining the carotid sinus pressure corresponding to 50% of maximum baroreceptor activity (EP_{50}) is illustrated.](http://ajpregu.physiology.org/)

\[\text{EP}_{50} = 60 \text{ mmHg}\]

\[\text{EP}_{59} = 90 \text{ mmHg}\]
To visually confirm the increased diameter of eNOS-transduced carotid arteries, we examined cross sections of unpressurized common carotid arteries and carotid sinuses from one rabbit. The diameter of carotid arteries subjected to eNOS gene transfer was larger than the diameter of the contralateral control arteries (Fig. 7).

**DISCUSSION**

The major findings of the present study are that adenoviral-mediated gene transfer of NOS to carotid sinus adventitia causes sustained NO-dependent inhibition of baroreceptor activity, a resetting of the baroreceptor pressure-activity curve to higher pressures, and an increase in carotid sinus diameter. The discussion addresses the possible mechanisms that may contribute to NOS-induced baroreceptor resetting, other factors to consider in interpreting the results, and the physiological and investigative implications of the findings.

**Possible Mechanisms of Baroreceptor Resetting Induced by NOS Gene Transfer**

Topical application of L-NAME to the carotid sinus prevented NOS-induced baroreceptor resetting (Fig. 4), strongly suggesting that the inhibition of baroreceptor activity was indeed dependent on NO production. The finding that topical application of adenoviral vectors to the carotid sinus led to localized transgene expression restricted to the adventitia layer of the vessel wall (Fig. 1) is in agreement with previous results (5, 26, 34).

**Cell source of transgene generated NO.** The predominant cells transduced by topical application of the AdeNOS vector to large arteries are adventitial fibroblasts (5). Previous studies have shown that adventitial cells expressing the NOS transgene, when stimulated by bradykinin or calcium ionophore, produce sufficient NO to provoke NO-dependent changes in vascular tone and structure in both endothelium-intact and endothelium-denuded arteries (5, 6, 31). Taken together, these results suggest that adventitial fibroblasts were the likely source of NO responsible for baroreceptor resetting in the NOS-transduced carotid sinuses. Generation of NO by eNOS is calcium dependent (9, 28). We speculate that calcium influx through mechanosensitive ion channels on fibroblasts (10, 37) may have activated eNOS and increased NO production in our experiments. We did not detect β-Gal transgene expression in the carotid sinus nerve (data not shown) consistent with the significant diffusion barrier presented by the perineurium surrounding nerve terminals in mature animals (21).

**Direct action of NO on baroreceptors.** NO may inhibit baroreceptor activity directly by an effect on baroreceptor nerve terminals. A previous study in our laboratory demonstrated that acute exposure of the isolated rabbit carotid sinus to NO or NO donors inhibits baroreceptor activity (25). The inhibition of nerve activity appeared to be mediated by a cGMP-independent mechanism, could not be explained by NO-induced vasodilation, and was transient, consistent with the short half-life of NO (25). In subsequent studies, we demonstrated that NO acutely inhibits voltage-depen-
dent Na⁺ currents in isolated baroreceptor neurons in culture (1, 23). The NO-induced inhibition of Na⁺ channels or channel-associated proteins (23). Thus NO may decrease baroreceptor activity by inhibiting Na⁺ channels that are essential for generation of action potentials. NO has also been shown to enhance activity of various K⁺ channels (2, 19) that conceivably could contribute to inhibition of baroreceptor activity. It is also possible that NOS-generated reactive oxygen species or peroxynitrite may inhibit baroreceptor activity (24).

Indirect action of NO via change in vascular compliance/structure. Alternatively, NO may decrease baroreceptor activity indirectly through changes in the compliance or structure of the blood vessel wall or by altering the coupling of baroreceptor terminals to the vessel wall. Arterial diameter of eNOS-transduced carotid sinuses was significantly larger than the diameter of β-Gal-transduced or control sinuses not exposed to adenovirus (Figs. 6 and 7). The increased diameter cannot be explained solely as a result of decreased vascular tone. Baseline tone is low in this preparation; acute administration of NO donors into the isolated carotid sinus region caused only a small increase in carotid sinus diameter (25). Furthermore, the increase in diameter of eNOS-transduced carotid sinuses was evident in unpressurized carotid arteries (Fig. 7), suggesting a structural enlargement that may be analogous to the structural remodeling that occurs in large arteries subjected to chronic increases in blood flow (17, 22, 36). Sustained increases in arterial flow increase vessel size (17, 22, 36), and increased flow or shear stress increases vascular eNOS expression (30, 35).
flow-induced arterial enlargement is attenuated by NOS inhibitors, suggesting that NO is a key mediator of the response (39). Enhanced NOS expression and NO production also mediate, at least in part, poststenotic dilatation of large arteries (4). Flow-induced changes in arterial structure (increased diameter) occur over a period of weeks to months (17, 22, 36). Our results suggest that NO-dependent arterial enlargement may occur as soon as 4–5 days after eNOS expression is enhanced. The mechanism may involve changes in the extracellular matrix (11, 12, 32, 36, 42). Although one might expect increased vessel diameter to increase baroreceptor activity, changes in the extracellular matrix may alter the mechanical coupling of the baroreceptor terminals to the vessel wall, leading to inhibition of activity despite the increase in carotid sinus diameter.

Possible role of arterial pressure in eNOS-induced baroreceptor resetting. The baroreceptor pressure-activity curve is reset to higher pressures in hypertensive states (20). Mean arterial pressure was not significantly different in rabbits subjected to β-Gal gene transfer (95 ± 2 mmHg) and eNOS gene transfer (93 ± 2 mmHg). Therefore, the resetting of the baroreceptor function curve in eNOS-transduced carotid sinuses cannot be explained by a difference in arterial pressure.

Other Factors for Consideration

The goal of the present study was to determine the effect of sustained overexpression of eNOS over a period of days on baroreceptor activity, thereby necessitating a comparison of activity measured in separate groups of control (β-Gal) and eNOS-transduced rabbits. The absolute level of recorded multifiber nerve activity is dependent on the recording conditions. Therefore, as is routinely done in these types of studies (24–26), we expressed baroreceptor activity as a percentage of maximum activity. This analysis allowed us to demonstrate a significant rightward shift of the pressure-activity relation to higher pressures for the eNOS-transduced carotid sinuses. The results do not enable an evaluation of a possible effect of NOS expression on absolute or maximum baroreceptor activity. Furthermore, we cannot rule out the possibility that selective damage to low-threshold baroreceptor fibers innervating the eNOS-transduced carotid sinuses might have contributed to the shift in the baroreceptor function curve to higher pressures.

Adenovirus may elicit an inflammatory response, particularly after intraluminal administration of adenoviral vectors to arteries that can limit the duration of transgene expression and alter physiological functions (29). In contrast, inflammation appears to be less of a problem after topical application of adenoviral vectors to adventitia of arteries (5, 26, 34). We have demonstrated previously that adenoviral-mediated gene transfer of β-Gal to carotid sinus adventitia did not significantly alter the baroreceptor pressure-activity relation (26), suggesting that adenovirus itself, β-Gal expression, and/or the potential inflammatory response did not alter baroreceptor sensitivity to a significant extent. The finding that the baroreceptor pressure-activity curve is shifted to higher pressures in eNOS-transduced vs. β-Gal-transduced carotid sinuses and the prevention of resetting by L-NAME suggest that the resetting was indeed caused by NOS overexpression and not by adenovirus or inflammation.

We evaluated the effect of eNOS gene transfer on baroreceptor activity 4–5 days after application of the viral vector to the carotid sinus. We chose this time point based on our previous study showing that transgene expression driven by the RSV promoter increased markedly between 1 and 4 days after application of the vector to carotid sinus adventitia (26). We observed that transgene expression is essentially absent ~1 mo after vector application (unpublished results). The loss of transgene expression over time may limit use of this technique to alter baroreflex sensitivity chronically over longer periods. Nevertheless, the results provide proof of principle that gene transfer can be used to chronically modulate baroreceptor function. Development of new vectors for gene transfer may extend the duration of transgene expression.

Perspectives

Endothelium, carotid sinus nerves, and fibroblasts contain eNOS, nNOS, and inducible NOS, respectively (9, 14, 28, 33, 40, 41), all of which can be upregulated by specific stimuli (9, 28). Thus there are several potential sources of NO located near, and possibly in, baroreceptor terminals. An inhibitory influence of endogenous NO on carotid chemoreceptor afferent activity that transgene expression driven by the RSV promoter increased markedly between 1 and 4 days after application of the vector to carotid sinus adventitia (26). We observed that transgene expression is essentially absent ~1 mo after vector application (unpublished results). The loss of transgene expression over time may limit use of this technique to alter baroreflex sensitivity chronically over longer periods. Nevertheless, the results provide proof of principle that gene transfer can be used to chronically modulate baroreceptor function. Development of new vectors for gene transfer may extend the duration of transgene expression.

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Gene transfer to sites of baroreceptor innervation potentially provides a method to address difficult, longstanding questions related to baroreceptor function. For example, by producing sustained changes in baroreceptor sensitivity, gene transfer experiments may provide new insight into the role of the baroreceptor reflex in long-term control of arterial pressure. In addition, gene transfer to carotid sinus and/or aortic
arch may provide a means to restore baroreceptor sensitivity in pathological states and examine the cardiovascular consequences of the improved reflex sensitivity. The approach may also provide a tool to explore the relation between chronic changes in vascular structure and changes in baroreceptor sensitivity.

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