Impaired Relaxation of the Carotid Artery During Activation of ATP-Sensitive Potassium Channels in Atherosclerotic Monkeys

Frank M. Faraci, PhD; Kristen Orgren, BA; Donald D. Heistad, MD

Background and Purpose This study examined the hypotheses that (1) atherosclerosis impairs relaxation of the carotid artery in response to activation of adenosine triphosphate (ATP)-sensitive potassium channels and (2) regression of atherosclerosis restores the response toward normal.

Methods Isometric tension was measured in rings of carotid artery taken from normal, atherosclerotic, and regression monkeys and precontracted submaximally with prostaglandin F2α.

Results Relaxation in response to acetylcholine was less in atherosclerotic compared with normal arteries (5±6% versus 54±4% [mean±SE] in response to 3x10⁻⁵ mol/L acetylcholine, P<.01). Relaxation in response to aprikalim, a direct activator of ATP-sensitive potassium channels, was also less in atherosclerotic than in normal arteries (32±7% versus 69±5% during 10⁻⁴ mol/L aprikalim, P<.01). Relaxation in response to aprikalim but not to acetylcholine or nitroprusside was inhibited almost completely by glibenclamide (4 μmol/L), a selective inhibitor of ATP-sensitive potassium channels. Relaxation in response to low but not high (10⁻⁴ to 10⁻³ mol/L) concentrations of sodium nitroprusside was less in atherosclerotic than in normal arteries. Regression of atherosclerosis tended to restore responses to acetylcholine, but not responses to nitroprusside or aprikalim, toward normal.

Conclusions These findings suggest that atherosclerosis impairs relaxation of the carotid artery in response to activation of ATP-sensitive potassium channels. Impaired responses may be due, in part, to nonspecific impairment of relaxation. Regression of atherosclerosis did not restore responses of the carotid artery toward normal. (Stroke. 1994;25:178-182.)

Key Words • acetylcholine • atherosclerosis • carotid arteries • monkeys • potassium channels
Sodium channels, produced marked relaxation of the carotid artery from normal monkeys (Fig 1). The highest intimal proliferation in this model. 16

Segments of carotid artery were cut into 5-mm rings and suspended in organ baths using two stainless-steel stirrups. A force transducer was used to record isometric tension in vessels continuously bathed with a modified Krebs solution with the following composition (in mmol/L): NaCl 118.3, KCl 4.7, CaCl2 2.5, MgSO4 1.2, KH2PO4 1.2, NaHCO3 25, ethylenediaminetetraacetic acid calcium 0.026, glucose 11.1, pH 7.4. The buffer was maintained at 37°C and aerated with 95% O2/5% CO2. Vessels were gradually stretched to a resting tension of approximately 2 g, which produced optimal contraction of approximately 80%. We did not examine the effects of higher concentrations of aprikalim because such experiments would have required relatively large quantities of aprikalim.

Glibenclamide, an inhibitor of ATP-sensitive potassium channels, and the DMSO vehicle had no significant effect on vascular tone in precontracted arteries. Glibenclamide produced almost complete inhibition of relaxation of the carotid artery in response to aprikalim (Fig 1). Aprikalim also produced relaxation in atherosclerotic arteries, but responses were significantly impaired compared with responses in normal arteries (Fig 1). For example, 10^-6 mol/L aprikalim produced relaxation in normal and atherosclerotic arteries by 69±5% and 32±7% (P<.01 versus normal), respectively. Regression of atherosclerosis was not associated with improvement of relaxation in response to aprikalim (P>.05) (Fig 1). As in normal arteries, relaxation in response to aprikalim in arteries from atherosclerotic and regression animals was inhibited by glibenclamide (Fig 1).

Acetylcholine, an endothelium-dependent vasodilator, produced relaxation of carotid arteries from normal animals that was not altered by glibenclamide (Fig 2). Relaxation in response to acetylcholine was significantly impaired in atherosclerotic arteries and was restored partially toward normal in arteries from regression animals (Fig 2).

Sodium nitroprusside, an endothelium-independent vasodilator, produced relaxation of normal arteries that was not affected by glibenclamide (Fig 3). Relaxation in response to nitroprusside tended to be less in atherosclerotic than in normal arteries (Fig 3). Responses to low concentrations of nitroprusside were inhibited significantly, but relaxation to higher concentrations of nitroprusside was not significantly different in normal and atherosclerotic blood vessels (Fig 3). Relaxation in response to nitroprusside was similar in carotid arteries.

Results

The carotid artery from normal monkeys had no gross or microscopic evidence of atherosclerotic lesions. In contrast, the carotid artery from atherosclerotic monkeys was observed to have lesions that ranged from fatty streaks to fibrofatty plaques. We have previously examined this artery morphometrically and observed marked intimal proliferation in this model. 16

Aprikalim, a direct activator of ATP-sensitive potassium channels, produced marked relaxation of the carotid artery from normal monkeys (Fig 1). The highest concentration of aprikalim (3x10^-6 mol/L) relaxed normal carotid arteries by approximately 80%. We did not examine the effects of higher concentrations of aprikalim because such experiments would have required relatively large quantities of aprikalim.
Discussion

There are two major findings in the present study. First, relaxation of the carotid artery in response to the endothelium-dependent agonist acetylcholine is impaired during atherosclerosis and tended to improve with dietary regression of atherosclerosis. These findings confirm previous studies describing impaired endothelium-dependent relaxation in other blood vessels during atherosclerosis. Relaxation of the carotid artery in response to acetylcholine was not altered by glibenclamide, suggesting that responses of this blood vessel to acetylcholine are not dependent on activation of ATP-sensitive potassium channels.

Second, activation of ATP-sensitive potassium channels with aprikalim produces glibenclamide-sensitive relaxation of the carotid artery. Thus, ATP-sensitive potassium channels appear to be present in the carotid artery in primates. Relaxation of the carotid artery in response to aprikalim was impaired significantly by atherosclerosis. Relaxation in response to low concentrations of nitroprusside was also impaired during atherosclerosis. Relaxation of atherosclerotic carotid arteries in response to nitroprusside or acetylcholine tended to be less than relaxation in normal arteries. This difference was statistically significant for low concentrations of nitroprusside. In contrast, maximum relaxation in response to higher concentrations of nitroprusside was not impaired significantly by atherosclerosis.

These findings in carotid arteries of monkeys, that atherosclerosis impairs relaxation in response to low but not high concentrations of a nitrovasodilator, are similar to findings in human coronary arteries. Sodium nitroprusside relaxes vascular muscle by activating guanylate cyclase, which produces an accumulation of cyclic guanosine monophosphate. The tendency for reduction of relaxation of atherosclerotic arteries in response to nitrovasodilators may be due to impairment in the guanylate cyclase system, or it may reflect structural alterations in the vessel wall. Nonspecific impairment of vasodilatation, if present, would presumably contribute to reduced relaxation in response to all agonists, but impairment of responses to aprikalim and acetylcholine in atherosclerotic vessels tended to be greater than impairment of responses to nitroprusside, especially at higher concentrations.

Responses to Acetylcholine

There is substantial evidence for endothelial dysfunction in atherosclerotic blood vessels. Atherosclerosis impairs basal and agonist-induced production or activity of EDRF and endothelium-dependent relaxation in humans and animal models of atherosclerosis. In agreement with this concept, we observed marked impairment of relaxation of the carotid artery in monkeys in response to acetylcholine, the "classic" agonist in studies of endothelium-dependent relaxation and EDRF. Relaxation of the carotid artery in response to acetylcholine was restored partially toward normal by dietary treatment of atherosclerosis. This observation supports previous studies suggesting that regression of atherosclerosis improves endothelium-dependent responses.

In some blood vessels, endothelium-dependent relaxation in response to acetylcholine appears to be mediated, in part, by release of an EDHF that activates ATP-sensitive potassium channels. Activation of these potassium channels produces hyperpolarization of vascular muscle and relaxation that is inhibited selectively by glibenclamide. In the present study, relaxation of the carotid artery in monkeys in response to acetylcholine was not altered by glibenclamide, which suggests that activation of ATP-sensitive potassium channels does not contribute to endothelium-dependent relaxation in response to acetylcholine in this artery. The concentration of glibenclamide used was efficacious because it produced marked inhibition of relaxation in response to aprikalim.

Responses to Sodium Nitroprusside

Relaxation of atherosclerotic carotid arteries in response to nitroprusside tended to be less than relaxation in normal arteries. This difference was statistically significant for low concentrations of nitroprusside. In contrast, maximum relaxation in response to higher concentrations of nitroprusside was not impaired significantly by atherosclerosis.

Fig. 3. Relaxation in response to sodium nitroprusside under control (Con) conditions (in the presence of vehicle) and in the presence of glibenclamide (Glib) in carotid arteries from normal (left panel), atherosclerotic (middle panel), and regression (right panel) monkeys. To examine responses to nitroprusside, arteries were precontracted submaximally with prostaglandin F2α. Values are mean±SE (n=9 normal, 6 atherosclerotic, and 7 regression animals). Relaxation in response to nitroprusside in the presence of vehicle was significantly reduced in the atherosclerotic (P<.05 vs normal at 3x10^-8, 10^-7, and 3x10^-7 mol/L nitroprusside) and regression (P<.05 vs normal at 3x10^-8, 10^-7, 3x10^-7, and 10^-6 mol/L nitroprusside) groups.

from atherosclerotic and regression animals (P>.05) (Fig 3).
apotassium channels. This impairment may be due to NS-24621, HL-14388, and AG-10269; research funds from the National Institutes of Health grants HL-38901, HL-16066, supply of aprikalim (RP52891). This study was supported by Investigator of the American Heart Association (92015170). Dr Faraci is an Established Investigator of the American Heart Association.

Acknowledgments

We thank Rhone-Poulenc Rorer (Cedex, France) for the supply of aprikalim (RF52891). This study was supported by National Institutes of Health grants HL-38901, HL-16066, NS-24621, HL-14388, and AG-10269; research funds from the Veterans Administration; and a Grant-In-Aid from the American Heart Association (92015170). Dr Faraci is an Established Investigator of the American Heart Association.

References

Impaired relaxation of the carotid artery during activation of ATP-sensitive potassium channels in atherosclerotic monkeys.
F M Faraci, K Orgren and D D Heistad

Stroke. 1994;25:178-182
doi: 10.1161/01.STR.25.1.178

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://stroke.ahajournals.org/content/25/1/178

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Stroke can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at: http://www.lww.com/reprints

Subscriptions: Information about subscribing to Stroke is online at: http://stroke.ahajournals.org//subscriptions/