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 (54±4% [mean±SE] in response to 3×10^{-4} 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^{-6} 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 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

See Editorial Comment, page 182

carotid artery in response to activation of ATP-sensitive potassium channels. We also determined whether dietary treatment of atherosclerosis (regression) restores responses to activation of ATP-sensitive potassium channels toward normal.

Materials and Methods

We studied three groups of adult cynomolgus monkeys. Nine normal monkeys were fed commercial chow (Purina Monkey Chow, Ralston Purina, Richmond, Ind), which produced plasma cholesterol concentrations of 119±6 (mean±SE) mg/dL. In a second group of six monkeys, atherosclerosis was induced by feeding the monkeys an atherogenic diet that contained 41% of total calories from fat and 0.8% cholesterol for 27±2 months. Plasma cholesterol concentrations in this group were 611±43 mg/dL. A third group of seven monkeys was fed an atherogenic diet for 21±1 months and was subsequently fed standard monkey chow for 16±4 months. The plasma cholesterol in this group was 694±57 mg/dL while they received the atherogenic diet and 117±8 mg/dL when they received the normal diet. We have described this primate model of regression of atherosclerosis and the morphological changes that occur in the carotid circulation in detail previously.16 Animals were sedated with 12 mg/kg ketamine and then anesthetized with 75 to 100 mg/kg α-chloralose intravenously. Supplemental anesthesia was administered as needed. The trachea was cannulated and the monkey ventilated mechanically with room air and supplemental oxygen. One common carotid artery was exposed, ligated proximally and distally with suture, and the isolated segment of artery was removed and placed into Krebs solution.
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 arteries and was restored partially toward normal in arteries from regression animals (Fig 2).

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).

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.

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.

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.
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 acetylcholine was not altered by glibenclamide, which suggests that aprikalim activates ATP-sensitive potassium channels. 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 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 vasodilation, 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 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.

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 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 vasodilation, 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 Aprikalim

Relaxation of the carotid artery in response to aprikalim was significantly impaired in atherosclerotic arteries. Relaxation of carotid arteries in response to aprikalim was inhibited almost completely by glibenclamide, which suggests that aprikalim activates ATP-sensitive potassium channels. Effects of glibenclamide were selective because the antagonist did not alter relaxation in response to nitroprusside or acetylcholine. These findings suggest that ATP-sensitive potassium channels are present and functional in the carotid artery of primates.

Potassium channels exist as a heterogeneous group including calcium-dependent and ATP-sensitive potassium channels. Two lines of evidence suggest strongly that aprikalim produces relaxation by activation of ATP-sensitive potassium channels. First, responses 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.
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 the American Heart Association (92015170). Dr Faraci is an Established Investigator of the American Heart Association.

In summary, the present study suggests that in addition to impairment of endothelium-dependent responses, atherosclerosis impairs relaxation of the carotid artery in response to activation of ATP-sensitive potassium channels. This impairment may be due to specific effects on ATP-sensitive potassium channels and nonspecific impairment of relaxation during atherosclerosis. Impairment of normal mechanisms that mediate vasodilatation may contribute to vasospasm during atherosclerosis.

Acknowledgments

We thank Rhone-Poulenc Rorer (Cedex, France) for the supply of aprikalim (RP52891). 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

doi: 10.1161/01.STR.25.1.178

*Stroke* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1994 American Heart Association, Inc. All rights reserved.
Print ISSN: 0039-2499. Online ISSN: 1524-4628

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/