Calcitonin gene–related peptide: a potent dilator of human epicardial coronary arteries

JEAN McEWAN, M.D., CH.B., SIMON LARKIN, B.Sc., GRAHAM DAVIES, M.D., SERGIO CHIERCHIA, M.D., MORRIS BROWN, M.D., JOHN STEVENSON, M.B., B.S., IAIN MACINTYRE, M.D., F.R.C.P., and ATTILIO MASERI, M.D., F.R.C.P.

ABSTRACT  To investigate the action of calcitonin gene–related peptide (CGRP) on human epicardial coronary arteries, six patients received intracoronary CGRP at doses of 50, 100, and 200 ng/min. The effect of CGRP was measured angiographically with a computerized analysis system. A dose-dependent increase in coronary arterial diameter was observed. At the highest dose there were 34%, 7%, 38%, and 40% mean increases in the diameters of the circumflex, proximal, mid, and distal left anterior descending arteries, respectively. No further increase in diameter was found after a subesequent dose of 1 mg intracoronary isosorbide dinitrate. Prior infusion of CGRP did not prevent coronary arterial spasm induced by ergonovine in two patients with variant angina, but a subsequent bolus of CGRP partially relieved the spasm. We propose that CGRP has a role in the regulation of coronary vascular smooth muscle tone.

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THE LOCALIZATION of neuropeptides in the cardiovascular system has suggested a physiologic role for peptidergic nerves in cardiovascular regulation. The existence of calcitonin gene–related peptide (CGRP) was predicted by analysis of the nucleotide sequence of the calcitonin gene and it was then identified in rat tissue, particularly within the cardiovascular system, by immunocytochemistry and immunoassay. Human CGRP was isolated from medullary carcinoma of the thyroid and characterized by fast atom-bombardment mapping. It has now been shown to be present in the plasma of normal humans and at plasma concentrations five times that of calcitonin, it is probably the major circulating product of the calcitonin gene. Studies both in vitro and in vivo have suggested that CGRP is a potent vasodilator and it appears to have a positive chronotrophic and inotropic effect on isolated rat heart auricle. To examine further its cardiovascular actions we studied the effect of the intracoronary infusion of CGRP on epicardial coronary arterial diameter and showed it to be a potent vasodilator.

Methods

The study was carried out with the approval of the Research and Ethical Committee of the Hammersmith Hospital. Eight patients were examined during routine coronary angiography. Seven were men, one was a woman, and their mean age was 52 years (range 45 to 58). Three patients had atypical chest pain and normal coronary arteries, two had variant angina with coronary arterial spasm superimposed on noncritical lesions, and three had effort angina and moderate stenosis of the mid left anterior descending (LAD) artery. In all patients selective coronary arteriography and left ventriculography were performed according to the Judkins technique. A No. 7F Goodale-Lubin catheter was positioned in the pulmonary artery by the percutaneous femoral vein approach. Urographin 76 was used as the contrast medium. After diagnostic coronary angiography, the most suitable view of the left coronary circulation was selected and the image intensifier was positioned; this position was maintained constant throughout the study. A control arteriogram was obtained in each patient. In six patients control vehicle solution (0.9% NaCl and 1% human serum albumin) followed by incremental doses of CGRP of 25, 50, and 100 ng/ml were infused at 2 ml/min into the left coronary artery for periods of 5 min. Human CGRP was the generous gift of Dr. Jan Pless, Sandoz Ltd.

After the control infusion and after each incremental dose of CGRP, blood pressure, pulmonary arterial pressure, and heart rate were recorded and coronary angiography was repeated. Two further patients received only control vehicle solution and underwent repeated arteriography. Three minutes after the final CGRP infusion four patients also received, by the intracoronary route, 1 mg isosorbide dinitrate (ISDN) and coronary angio-
The effects of CGRP on ergonovine-induced coronary spasm were evaluated in two patients with variant angina. After the cessation of the infusion of the maximal dose of CGRP, intravenous ergonovine was administered in incremental doses of 0.025, 0.05, and 0.1 mg at intervals of 4 min. In each subject provocative testing before catheterization had resulted in electrocardiographic changes and pain suggestive of coronary spasm at this cumulative dose. In both patients, coronary spasm with chest pain and ST segment elevation was again induced and was now confirmed angiographically. In one patient the spasm was immediately reversed by 2 mg intracoronary ISDN, but in the second patient a bolus dose of 1.5 µg CGRP was given in an attempt to reverse spasm and the left coronary artery was again visualized.

**Quantitative angiographic analysis system.** In each patient coronary arterial diameter was measured at specific distances from identifiable branching points in end-diastolic frames after control or CGRP infusions and, where appropriate, after 1 mg intracoronary ISDN.

Quantitative analysis of selective coronary arterial segments was carried out with the help of an automatic computer-based coronary angiography analysis system (CAAS-Pie Data), as described elsewhere. 11-13 End-diastolic frames from each arteriogram were selected by one observer (J. M.) and analyzed in random order for each patient by an independent “blinded” observer (S. L.). A selected coronary arterial segment was defined manually by a number of centerline positions along that segment. The regions of interest were digitized and stored on a LSI 11/73 minicomputer and the contour positions of the segment were detected automatically on the basis of signal-density first and second derivative criteria. With the use of the contour data the diameter function of the segment was computed. The angiographic catheter was used as a scaling device and this, together with pincushion distortion correction, allowed the diameters to be recorded as absolute values (mm).

Measurements of epicardial arterial diameters were made at four sites: at the proximal, mid, and distal LAD, and at the proximal circumflex arteries. Routine angiography showed, in one patient, a short left main stem and poor visualization of the circumflex artery, so no measurement of the circumflex arterial diameter was made in this subject. Statistical analysis was by two-way analysis of variance with replication or by paired t test.

**Results**

Intracoronary CGRP caused a dose-dependent increase in epicardial coronary arterial diameter (figures 1, 2, and 3). At 200 ng/min, the mean increases in diameter were 35%, 7%, 38%, and 40% in the circumflex, proximal, and mid and distal LAD arteries, respectively. No further vasodilation was produced by 1 mg intraocoronary ISDN given 3 min after the cessation of the infusion of CGRP (figure 4).

In the two patients with variant angina, there were vessel wall irregularities but no significant lesions at the site at which spasm was induced. In these patients, although both the normal artery and the site at which spasm occurred in the proximal LAD segment increased in diameter after the infusion of CGRP, the electrocardiographic changes, pain, and vasospasm induced by ergonovine were not prevented by the prior infusion of CGRP. Only at that site at which spasm occurred was there a profound decrease in the coronary arterial diameter after ergonovine that resulted in subtotal occlusion of the vessel. Analysis of the response of a further distinct segment of angiographically normal artery in the circumflex artery to both CGRP and ergonovine showed relaxation after CGRP and constriction after ergonovine, but no spasm induction. In
one patient a 1 µg intracoronary bolus of CGRP partly relieved the spasm and its associated features (figure 5).

No significant change in epicardial coronary arterial diameter was observed in the subjects receiving only control vehicle solution.

The only systemic effect noted at the doses of CGRP given was mild facial flushing in three patients. No change in heart rate or systemic or pulmonary arterial pressure was observed.

**Discussion**

The demonstration of significant coronary vasodilation by CGRP adds weight to the suggestion of the existence of a peptidergic system that controls vascular smooth muscle tone.

Studies in vitro of rat and human CGRP showed

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**FIGURE 3.** Percentage change in coronary arterial diameter (mean ± SEM) with incremental 5 min doses of CGRP. Significant increases in diameter occurred with increasing doses of CGRP in all segments, but the increase was greater in segments of smaller diameter than in those of larger diameter. Circumflex, p < .05; proximal LAD, p < .05; mid LAD, p < .001; proximal LAD, p < .01 (analysis of variance).
Substance P, which is a vasodilator, and for NPY, which is a vasoconstrictor, but CGRP is probably the most potent endogenous vasodilator that has been studied in humans. The amount of CGRP required to produce vasodilation is low. In this study we gave a dose appropriate to the coronary circulation: our maximum dose was one-twentieth of that reported to be given intravenously. No further dilatation of the coronary arteries was observed when intracoronary ISDN was given a few minutes after cessation of the CGRP infusion, despite the fact that the dose of ISDN was 56.4 nmol/kg while the cumulative dose of CGRP given was only 6.65 pmol/kg. The short delay between the cessation of the infusion of the maximum dose of CGRP and the administration of ergonovine should not have affected the results since venous occlusion plethysmography of the forearm during the brachial arterial infusion of CGRP has demonstrated a biological half-life of 18 min.

The inability to prevent focal spasm induced by ergonovine suggests that CGRP is unlikely to be involved pathophysiologically in variant angina. Patients with variant angina exhibit a marked localized hyperreactivity to a variety of vasoconstrictor stimuli that is well beyond the physiologic range of response; in the two patients with variant angina in our study there was no evidence that CGRP could reverse this hyperreactivity. It will, however, be necessary to examine the effects of CGRP in patients with other forms of angina in whom less dramatic changes in coronary vasomotor tone are likely to play a pathophysiologic role.

The heart is supplied with both sympathetic and parasympathetic nerves and the coronary arteries have both vasoconstrictor $\alpha$-29 and vasodilatory $\beta$-adrenergic receptors; parasympathetically mediated vasodilation can also be demonstrated.31 Colocalization of neuropeptides with classic neurotransmitters, e.g., NPY with norepinephrine and CGRP with acetycholine,33 suggests that these peptides may be neuromodulators and an integral part of the neural control of vascular smooth muscle tone. This is supported by the demonstration of potentiation of adrenergic vasoconstriction by NPY.34

The control of coronary smooth muscle tone is as yet poorly understood, but the present study supports neuropeptide involvement in its regulation.

References
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