HISTAMINE acts through at least two specific receptors.\(^1\)\(^2\) The development of \(H_1\) receptor antagonists\(^3\) and more recently of specific \(H_2\) antagonists\(^4\) has made it possible to identify the presence in tissues of these two receptors. Histamine is taken up actively by the heart\(^4\) and its release may be quantified.\(^5\)\(^6\) But the role of histamine in the regulation of the coronary circulation and the nature of coronary histamine receptors are unknown.

In the present investigation we have studied the action of histamine in the coronary circulation, using low dose infusions of histamine and specific \(H_1\) and \(H_2\) receptor antagonists. By giving drugs in this way we avoided systemic effects. The results show that there are \(H_1\) and \(H_2\) receptors in the coronary vasculature with vasodilator effects similar in magnitude to that found during reactive hyperemia following brief coronary artery occlusions. We extended the study to examine the effects on myocardial reactive hyperemia of blocking histamine responses with both \(H_1\) and \(H_2\) receptor antagonists.

**Methods**

**EXPERIMENTAL MODEL**

Experiments were performed on anaesthetised, healthy, young greyhounds weighing 19-30 kg. Anaesthesia was induced with thiopentone (20 mg/kg, iv) and maintained by ventilation with a Harvard respirator using a 2:1 nitrous oxide-oxygen mixture and sodium pentobarbital. Sodium pentobarbital (20 mg/kg, iv) was given as an initial bolus and further doses were administered as necessary to suppress respiration. The nitrous oxide-oxygen gas mixture was passed through a humidifier to maintain body temperature at approximately 37.0°C.\(^7\) The respiratory rate and gas mixture were adjusted as necessary during the experiment to maintain blood gases and pH at physiological values.

Through a left thoractomy, the pericardium was incised and the heart suspended in a pericardial cradle. The circumflex branch of the left coronary artery was dissected free for approximately 2 cm where there were no major branches. An appropriately sized (inside diameter (i.d.). 3-4 mm) IVM electromagnetic flow transducer (model FT245S) was placed around the exposed artery. A pneumatic occluder with an inflatable cuff was placed distal to the flow transducer. A polyvinyl catheter (i.d., 0.4 mm; outside diameter (o.d.) 0.8 mm) was inserted through the circumflex coronary artery wall by a modification of the Herd and Barger technique\(^8\) to allow close arterial infusions of drugs. The catheter was passed in and out through the arterial wall and the catheter end was drawn through for approximately 30 cm. Both ends of the catheter were cut to equal length and infusions were passed through a small side hole in the intraluminal portion of the catheter. This allowed simultaneous infusions of two drugs. A polyvinyl catheter (1 mm. i.d.; 2 mm. o.d.) was passed into the abdominal aorta via the femoral artery for arterial blood pressure measurements. Coronary blood flow (CBF) was measured with an EMI electromagnetic flowmeter (SFMB-1 type 28). The signal to noise voltage ratio was 70:1. baseline drift less than 2%. and calibration accuracy.
Percent repayment of flow debt
response. Flow debt, reactive hyperemic flow, and percent
compared only if resting CBF. blood pressure, and heart
rate values remained within ±10% throughout the experi-
ment. At least 5 minutes were allowed between each
measurement. Reactive hyperemic flow was obtained by planimetry. Reactive hyper-
emia was produced by inflating the pneumatic cuff to produce zero flow and then
deflating it after 8 seconds. Changes in CBF. blood pressure, and heart rate were recorded until CBF had returned to preocclusion values.

Reactive hyperemic flow was obtained by rapidly inflating the pneumatic cuff to produce zero flow and then rapidly deflating it after 8 seconds. Changes in CBF. blood pressure, and heart rate were recorded until CBF had returned to preocclusion values.

Reactive hyperemia following 8-second occlusions and flow responses to drug infusions were recorded at a paper speed of 2.5 mm/sec with electrical means of CBF and pressure. The integral of the flow curve following reactive hyperemia was obtained by planimetry. Reactive hyperemic responses and flow responses to drug infusions were compared only if resting CBF. blood pressure, and heart rate values remained within ±10% throughout the experiment. At least 5 minutes were allowed between each occlusion or infusion to allow complete recovery of the response. Flow debt. reactive hyperemic flow, and percent repayment of flow debt were calculated as described by Coffman and Gregg11 according to the following formula:

Flow debt = control flow rate x duration of occlusion

Reactive hyperemic flow
= (integral of the flow curve during reactive hyperemia
– (control flow rate x duration of reactive hyperemia)

Percent repayment of flow debt
= reactive hyperemic flow/flow debt

In analysis of results. Student's t-test was used to deter-
mine significance of differences between paired observa-
tions in each group of experiments. Results are expressed as means ± se.

Results
In the 16 dogs studied. CBF was 81 ± 6 ml/min (range.
35-126 ml/min). Blood pressure was 130 ± 5 mm Hg (range.
95-152 mm Hg), and heart rate 183 ± 4 beats/min (range.
150-198 beats/min).

HISTAMINE
The doses of histamine used in this study were those
found in preliminary studies to produce a dose-related
response in the coronary circulation without producing systemic effects. Figure 1 is a representative experiment showing CBF responses to 1-minute intracoronary infusions of 5. 10, and 20 /ig/min of histamine. CBF increase began about 20 seconds after the infusion commenced and reached a peak value at about 30 seconds. By allowing for the expected catheter transit time of 16 seconds. the CBF response to histamine occurred about 4 seconds after his-
tamine had actually reached the coronary artery. In re-
sponse to a histamine infusion of 5 /ig/min. CBF rose from
a resting value of 76 ml/min to a peak of 172 ml/min. an
increase of 126%. The CBF returned rapidly to resting
values on cessation of the histamine infusion. In response
to a histamine infusion of 10 /ig/min. the CBF rose from
72 ml/min to a peak of 213 ml/min. an increase of 196%. When histamine was infused at 20 /ig/min. the peak in-
crease was 253%. During each infusion. heart rate and
blood pressure did not change. Following each infusion. CBF returned to resting values and a 5-minute interval
was allowed before any further infusion was performed.
An infusion of saline (0.9%) did not alter CBF. heart rate.
or blood pressure.

To assess reproducibility of histamine responses. in
three separate dogs infusions of histamine only at 5. 10.
and 20 /ig/min were given for the same number and
duration of infusions used in histamine-blocking experi-
ments. There was very little variation in five consecutive
dose-response curves in each dog. with no evidence of
tachyphylaxis to histamine. Histamine dose-response
curves were performed twice in each preparation prior to
the infusion of histamine blockers; there was no variation in
responses in each dog.

METIAMIDE AND MEPYRAMINE
In preliminary experiments. mepyramine (100 /ig/min)
or metiamide (100 /ig/min) given separately by intracoro-
nary infusion for 3 minutes did not affect heart rate, blood pressure, or CBF. When mepyramine and metiamide were given together for 3 minutes there was a small but insignificant reduction in CBF [4.5 ± 1% (mean ± SE) in the 12 dogs studied]; but there was no change in heart rate or blood pressure. When mepyramine or metiamide were given at the dose of 1,000 μg/min for 3 minutes in separate dogs there was no significant increase in the blockade of the CBF response to histamine (20 μg/min) given by intracoronary infusion. With these large doses the preparations tended to deteriorate rapidly, therefore the lower doses of 100 μg/min of both mepyramine and metiamide were used.

HISTAMINE AND METIAMIDE

Figure 2 is a representative experiment showing the response to histamine (20 μg/min) given before and after a 3-minute infusion of metiamide. The metiamide infusion was commenced 2 minutes before histamine was given and continued during the 1-minute histamine infusion. Histamine increased CBF from 92 ml/min to a peak of 253 ml/min, an increase of 175%. Histamine (20 μg/min) in the presence of metiamide increased the flow by 103%. Finally, histamine (20 μg/min) in the presence of both metiamide and mepyramine increased flow by only 9%

The group data showing the effects of histamine blockade in six dogs are summarized in Figure 3. CBF was 73 ± 12 ml/min (range, 35–104 ml/min) in these dogs. After metiamide there was a significant parallel displacement of the dose-response curve to the right (P < 0.05, paired observations) with a dose ratio of 2. Also seen in Figure 3 is the effect of combined blockade by metiamide and mepyramine after metiamide alone. The dose-response curve was shifted further to the right with a dose ratio of 16. At the highest dose of histamine (320 μg/min) used after simultaneous mepyramine and metiamide infusions, there were systemic effects. However, the small decrease in blood pressure and increase in heart rate occurred after the infusion of histamine had ceased, and recovered to normal values within 1 minute.

HISTAMINE AND MEPYRAMINE

Mepyramine (100 μg/min for 3 minutes) reduced the CBF response to histamine. A representative experiment is shown in Figure 4. Histamine (20 μg/min) increased CBF from 66 ml/min to a maximum flow of 255 ml/min. a

286% increase. When mepyramine (100 μg/min) was given 2 minutes before histamine and continued during the histamine infusion, histamine increased CBF by 63%. Finally, in the presence of both mepyramine and metiamide, the histamine response was almost abolished. There was only a 4% increase in flow.

The group data in six dogs are summarized in Figure 5. CBF was 81 ± 11 ml/min (range, 67–126 ml/min) in these dogs. There was a parallel significant (P < 0.01, paired observations) shift to the right of the histamine dose-response curve (dose ratio = 4). In the presence of combined mepyramine and metiamide blockade there was a further parallel shift to the right of the dose-response curve (dose ratio = 16). These combined histamine blockade responses are similar to those seen in the six dogs in which metiamide blockade was given initially (Fig. 3).

CONTRACTILITY CHANGES IN RESPONSE TO HISTAMINE

In two dogs left ventricular contractility during infusions was assessed using peak rate of change of left ventricular pressure (dP/dt). In the resting state, peak dP/dt measured for 10 consecutive beats was 1,749 ± 23 mm Hg/sec; during the histamine (20 μg/min) dP/dt was unchanged at 1,743 ± 21 mm Hg/sec. Likewise there were no contractility changes in response to histamine (40 μg/min) in the presence of mepyramine (100 μg/min) or to histamine
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amide (100 μg/min). CBF = coronary blood flow. Values are means ± SE.

Table 1. Results (of five experiments in four dogs) are expressed as mean ± SE.

<table>
<thead>
<tr>
<th>RH (% repayment)</th>
<th>Peak flow (% increase)</th>
<th>Duration of RH (sec)</th>
<th>Heart rate (beats/min)</th>
<th>Blood pressure (mm Hg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before histamine blockade</td>
<td>616 ± 120</td>
<td>175 ± 27</td>
<td>87 ± 9</td>
<td>171 ± 9</td>
</tr>
<tr>
<td>After histamine blockade</td>
<td>620 ± 91</td>
<td>177 ± 19</td>
<td>83 ± 11</td>
<td>171 ± 9</td>
</tr>
</tbody>
</table>

Discussion

The present study shows that histamine is a potent coronary vasodilator. Peak flow responses to close coronary arterial infusions of histamine without systemic effects are similar to those seen with intracoronary adenosine infusions.22–25 The maximum increase in CBF with histamine was similar to the increase with reactive hyperemia following brief coronary occlusions, and the latter probably represents maximal coronary vasodilation. By using a coronary arterial catheter, we were able to give low dose infusions which reached the vascular receptors without producing systemic effects. In the two dogs in which contractility changes were assessed, there was no alteration in contractility to the dose of histamine used by itself or in combination with the histamine antagonists. Thus, CBF responses were due only to the direct action of the infused drugs and not to metabolic changes secondary to contractility changes.

Powell and Brody14 recently described positive chronotropic and negative inotropic cardiac responses to intravenous histamine. These were not seen in our study with histamine and its antagonists given selectively into the circumflex branch of the left coronary artery. Thus we were able to avoid systemic effects which could produce reflex changes in the heart rate or contractility. Also, by selecting a site on the coronary artery distal to branches supplying the atria, we were able to avoid direct effects on pacemaking and conducting tissue in the heart. Although histamine was infused only into the circumflex artery, this supplies most of the free wall of the left ventricle so that contractility changes would have been recorded had there been significant inotropic effects.

The mean CBF in the present study (79 ± 11 ml/min) would have resulted in histamine concentrations of approximately 0.05 μg/ml, 0.15 μg/ml, and 0.25 μg/ml, respectively, with infusions of 5, 10, and 20 μg/min. Anrep and associates,15 using dog heart-lung preparations in which histamine was injected at a concentration of 0.1 μg/ml of blood, found CBF increases similar to those reported here.

When histamine antagonists were given singly or in combination, dose-response curves were shifted to the right in a parallel manner, indicating competitive inhibition. Given alone, mepyramine and metiamide did not change CBF, heart rate, or blood pressure. Given in combination, these drugs decreased CBF slightly but did not change heart rate or blood pressure. Thus, changes in CBF response to histamine in the presence of the histamine antagonists singly or in combination are due to their blocking action and not to an independent action of the antagonists on the coronary vascular receptors. There was no attenuation in histamine responses during the course of an experiment, indicating absence of tachyphylaxis. Likewise, with the low doses of drugs, there was no evidence of deterioration of the preparation as reported by Parsons and Owen.16

Dose ratios of histamine were compared at 50% of the maximum response found in the control dose-response curves. The dose ratios of histamine in the presence of
Histamine receptors in the coronary circulation/Giles et al.

Histamine depressor responses have been reported in the cat and were partially blocked by mepyramine and abolished with the addition of burimamide or metiamide; yet H₂ blocking drugs alone had no effect on the histamine depressor response.¹⁰ Our results show that H₁ receptor activity appears to predominate in the coronary vasculature. The doses of mepyramine and metiamide used gave near-maximal blocking effects without affecting resting CBF, heart rate, or blood pressure. The H₁ and H₂ receptors appear to have separate activities and histamine analogues have either H₁ or H₂ activity.¹⁻² The H₁ receptor activity predominates and the augmented histamine blocking effect seen when mepyramine and metiamide are given in combination could be interpreted by either of the two models suggested by Powell and Brody.¹⁷ In the first model they suggest that histamine activates both H₁ and H₂ receptors, yet H₁ activity predominates, which could be due to (1) a greater number of H₁ receptors, (2) a greater affinity of histamine for H₁ receptors, or (3) greater smooth muscle action as a result of H₁ receptor activation. The second model suggests that histamine activates both H₁ and H₂ receptors equally, and if one of the group of receptors is blocked the action of the other is enhanced as a result of more histamine being made available to the remaining receptors. Our data do not indicate which of these possible interactions has occurred.

A physiological role for histamine in the regulation of blood flow has not been defined. Anerup and associates¹⁵,¹⁰ described the release of histamine-like substance from the dog heart and exercising human forearm and attributed reactive hyperemia to the production of histamine. Others²¹⁻²⁵ were unable to confirm these findings and questioned the method used to detect histamine in humans. Duff and associates²⁸ found that H₂ blocking drugs, including mepyramine, reduced reactive hyperemia following prolonged occlusions (10-15 minutes) of forearm blood flow, but had no effect on shorter occlusions (3-5 minutes). In the dog heart, we found that close coronary infusion of histamine produced vasodilation similar to that seen in reactive hyperemia following brief occlusions and that this histamine response could be abolished by H₁ and H₂ receptor antagonists given in combination. However, in the presence of both histamine antagonists there was no change in the reactive hyperemia. Folkow and associates²⁹ found that the classical H₁ antihistamines did not affect reactive hyperemia. They also suggested that there was a second histamine receptor that these antihistamines did not block. Our studies show that effective H₁ and H₂ receptor blockade does not change reactive hyperemia following brief coronary artery occlusions.

The use of intracoronary histamine antagonists to identify histamine receptors assumes that these drugs are able to reach all such receptors. With intra-arterial histamine infusions, this appears to be so as the histamine effect on vascular receptors is blocked in the doses used. Moreover, increasing the doses of antagonists 10-fold did not alter the degree of blockade. We cannot exclude the possibility that what Dale³⁰ has referred to as "intrinsic histamine" is released in response to brief occlusions and that the histamine antagonists are unable to block this effect. However, this is probably unlikely as it has been shown that histamine stimulates adenyl cyclase and produces cyclic AMP, effects which the H₂ blocker burimamide competitively inhibits in both the intact heart³¹ and in homogenates of myocardium.³²

Our data show there are H₁ and H₂ histamine receptors in the coronary vasculature of the dog and that histamine is a coronary vasodilator. The histamine receptors are competitively blocked with specific H₁ and H₂ antagonists and, when given together, each antagonist augments the effect of the other. Although the role of histamine in the regulation of coronary blood flow remains unknown, histamine activity does not appear to be a factor in reactive hyperemia following brief coronary artery occlusions.

Acknowledgments

We thank Patrick Legge for technical assistance and Pat Wilde for secretarial assistance.

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A Method for Determining Segmental Resistances in the Microcirculation from Pressure-Flow Measurements

STEPHEN H. NELLIS AND BENJAMIN W. ZWEIFACH

SUMMARY On the basis of an electrical analog, open circuit impedance functions were used to analyze the microcirculation. No specific structure need be assumed except a two-port, two-terminal network in which the major artery and vein supplying the tissue represent the input port and the two ends of the microvessel under study are the output port. The open circuit measurements were made by occluding microvessels in the exteriorized omental bed of anesthetized rabbits. The pressure upstream to the occlusion defines the source pressure of a Thévenin's equivalent circuit. The equivalent resistance value was calculated by plotting the flow through a given microvessel against the pressure developed during a gradual occlusion. The changes in pressure vs. the changes in flow during a progressive occlusion were found to be linearly related. The Thévenin's equivalent resistance was maximum downstream to an occluded artery and upstream to the occluded vein. Within the capillary network, source pressures consistently were within a narrow range. Topically applied norepinephrine resulted in marked changes in source resistance and no changes in source pressures. Threshold doses of norepinephrine given intravenously resulted in changes in source pressures, but minimal changes in source resistance, even though a substantial change in vascular resistance was indicated when calculated on the basis of arterial pressure minus micropressure divided by microvessel flow. The present method defines the functional characteristics of the distributing vessels in terms of two pressures and two equivalent resistances and is relatively easy to perform. The technique can be used to determine the vascular components involved in the response to particular stimuli.

PRESSURE-FLOW relationships across discrete organs or masses of tissue usually are described by a lumped numerical expression representing the effective resistance encountered by the blood during its passage from artery to vein. A more precise description of the functional organization of the system would be feasible if the resistance in specific segments of the vascular tree could be measured. Pressure and flow interactions at the tissue level are difficult to formulate because of the complex structural organization of the microvascular network. An especially formidable difficulty stems from the fact that discrete segments of the microcirculation are not supplied by a single source, nor are they drained by one vessel. In addition, the microvessels interconnect extensively at all levels of the system. Methods that do not involve a detailed analysis of all of the vascular elements must define alternative procedures for characterizing a network by a single peripheral resistance value. A possibility in this regard is to make certain a priori assumptions about the characteristics of the network bed. One such assumption is that it be represented by a simple parallel group of resistance vessels.

During the past few years, because of the capability of obtaining large numbers of direct measurements of microvascular indices, it has become possible to set up well documented distribution plots of both pressure and flow for discrete anatomical segments of a particular network. Such average values have been used to depict the spatial distribution of vascular resistance on the basis of idealized...
Histamine receptors in the coronary circulation of the dog. Effects of mepyramine and metiamide on responses to histamine infusions.

R W Giles, G Heise and D E Wilcken

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