Abstract

Anaphylactic events occurring in cardiac tissues can result in cardiac dysfunction via vasoconstriction and arrhythmias. Calcitonin gene-related peptide (CGRP) is the most potent vasodilator and possesses anti-arrhythmic action. We examined the influence of CGRP on cardiac anaphylaxis in guinea-pigs. In the Langendorff-perfused heart of passively sensitized guinea-pigs, antigen challenge evoked a decrease in coronary flow, left ventricular pressure and its maximum first derivatives (±dP/dtmax) and an increased heart rate. Antigen challenge also induced atrioventricular conduction block. Treatment with CGRP (1 or 3 nM) significantly improved the recovery of cardiac function and reduced the incidence and duration of atrioventricular block without influencing the increased heart rate. Pretreatment with capsaicin caused effects similar to those of CGRP and markedly elevated the content of CGRP in coronary effluent. Ischaemic preconditioning, induced by two cycles each of 5 min global ischaemia and 5 min reperfusion, also improved cardiac function and raised the level of CGRP in coronary effluent. The protective effects of ischaemic preconditioning were abolished in the presence of the CGRP receptor antagonist CGRP 8–37. Histamine release did not differ significantly during any of the interventions. The findings of the present study indicate that, in guinea-pig hearts, CGRP protects against cardiac anaphylaxis and that the cardioprotection by CGRP is independent of histamine release.

Key words

Calcitonin gene-related peptide · Cardiac anaphylaxis · Cardiac function

Introduction

Acute interaction of antigen with specific antibodies present in cardiac tissues results in rapid cardiac dysfunction, including an increased heart rate, coronary vasoconstriction and arrhythmias. This immediate hypersensitive reaction is termed cardiac anaphylaxis (Cooper 1993). Mediators play a vital role in cardiac anaphylaxis. The release of histamine from cardiac mast cells can provoke tachyarrhythmia (Marone et al. 1995). Leukotrienes, platelet-activating factor or thromboxane contribute to the enhancement of coronary vascular resistance (Mest et al. 1995). Accumulation of adenosine reportedly disturbs the atrioventricular nodal conduction (Hughes et al. 1984). Previous investigations have shown that bradykinin and angiotensin converting enzyme inhibitors, which stimulate bradykinin production, attenuate the cardiac anaphylactic reaction, and it has been suggested that the effect of bradykinin may be due to opposing the vasoconstricting effects of some anaphylactic mediators (Rubin and Levi 1995).

Calcitonin gene-related peptide (CGRP), a principal neurotransmitter of capsaicin-sensitive cardiac sensory nerves, has potent vasodilator effects (Brain et al. 1985; Bell and McDermott 1996). Pretreatment with CGRP has been shown to protect the myocardium against injury by a variety of harmful factors (Li et al. 1996; Peng et al. 1996, 1998) and it has been suggested that the cardioprotection afforded by ischaemic preconditioning is related to stimulation of endogenous CGRP release (Li et al. 1996; Fardinandy et al. 1997). Therefore, in the present study, we examined the protective role of endogenous and exogenous CGRP in cardiac anaphylaxis in the guinea-pig.

Materials and methods

Isolated heart perfusion. Male guinea-pigs weighting 300–350 g were anaesthetized with pentobarbitone sodium (40 mg/kg, i.p.). Hearts were excised rapidly and perfused retrogradely at a constant perfusion pressure of 100 cm H2O in a Langendorff apparatus with Krebs-Henseleit (K-H) buffer saturated with 95% O2 and 5% CO2. The K-H buffer solution had the following composition (in mM):

- NaCl: 118.2
- KCl: 4.7
- CaCl2: 2.5
- MgSO4: 1.2
- NaH2PO4: 1.2
- NaHCO3: 25.2
- Na2HPO4: 1.2
- glucose: 11.5
- pH: 7.4

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Isolated heart anaphylaxis. Male guinea-pigs were passively sensitized by an i.p. injection of the three successive doses of 5 mg bovine serum albumin every alternate day. Three weeks after the last injection, the hearts of the sensitized guinea-pigs were isolated and perfused as above and eventually challenged intra-aortically with 5 mg bovine serum albumin in 0.2 ml K-H buffer.

CGRP assay. The whole coronary effluent of perfusion fraction (5 min) was collected before and after ischaemia or treatment with CGRP, capsaicin, and acetic acid (final concentration 0.2 mM) was added. (5 min) was collected before and after ischaemia or treatment with CGRP. CGRP-like immunoreactivity (CGRP-LI) in the perfusion before antigen injection. In the case of ischaemic preconditioning, hearts were perfused with CGRP 8–37 (100 nM) for 5 min, and then challenged by bovine serum albumin. For antigen-challenged group, sensitized guinea pig-hearts were challenged with specific antigen resulted in a cardiac anaphylactic reaction, as shown by a decrease of CF, LVP, ±dP/dt_max and an increase of HR. CGRP (1 or 3 nM) caused a significant improvement of cardiac function (P<0.05) (Table 1, Fig. 1).

To examine cardioprotection elicited by endogenous CGRP, we exposed the myocardium to capsaicin, which selectively stimulates the release of transmitters from sensory nerves. Pretreatment with capsaicin (100 nM) caused a significant increase of CGRP release in coronary effluent concomitantly with an improvement of cardiac function (P<0.05) (Table 1, Figs. 1 and 2)

Ischaemic preconditioning induced by two cycles of 5 min ischaemia and 5 min reperfusion also attenuated the anaphylactic reaction of hearts challenged by antigen, as shown by improvement of the recovery of cardiac function (P<0.05) (Table 1, Fig. 1). The content of CGRP in coronary effluent was significantly increased during ischaemic preconditioning (Fig. 2). To explore the possible contribution of endogenous CGRP, we used CGRP<sub>8–37</sub>, the CGRP receptor antagonist. The cardioprotection afforded by ischaemic preconditioning was abolished in the presence of CGRP<sub>8–37</sub>.

Effects of CGRP, capsaicin or ischaemic preconditioning on cardiac function

Results

Effects of CGRP, capsaicin or ischaemic preconditioning on cardiac function

Sensitized guinea pig-hearts challenged with specific antigen resulted in a cardiac anaphylactic reaction, as shown by a decrease of CF, LVP, ±dP/dt_max and an increase of HR. CGRP (1 or 3 nM) caused a significant improvement of cardiac function (P<0.05) (Table 1, Fig. 1).

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Effects of CGRP, capsaicin or ischaemic preconditioning on HR and rhythmicity

Antigen challenge evoked tachycardia and A-V conduction block (Fig. 1, Table 2). CGRP, capsaicin or ischaemic preconditioning significantly decreased the incidence and duration of A-V conduction block (Table 2), but did not slow the tachycardia induced by antigen challenge (Fig. 1).

### Table 1 Effect of calcitonin gene-related peptide (CGRP), capsaicin, chlorpheniramine or ischaemic preconditioning (PC) with or without CGRP receptor antagonist (CGRP<sub>8–37</sub>) on coronary flow. Means±SEM, n=10–12

<table>
<thead>
<tr>
<th></th>
<th>Pre-antigen</th>
<th>Post-antigen (min)</th>
<th>1</th>
<th>3</th>
<th>5</th>
<th>11</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antigen</td>
<td>15.0±0.8</td>
<td>6.8±0.6</td>
<td>6.8±0.4</td>
<td>7.3±0.4</td>
<td>7.4±0.4</td>
<td></td>
</tr>
<tr>
<td>+CGRP (1 nM)</td>
<td>14.9±1.4</td>
<td>9.5±0.8*</td>
<td>9.7±0.6**</td>
<td>10.4±0.6**</td>
<td>11.5±0.7**</td>
<td></td>
</tr>
<tr>
<td>+CGRP (3 nM)</td>
<td>14.7±1.3</td>
<td>11.1±0.7**</td>
<td>11.0±0.5**</td>
<td>10.7±0.5**</td>
<td>13.2±0.8**</td>
<td></td>
</tr>
<tr>
<td>+capsaicin</td>
<td>14.3±1.2</td>
<td>10.0±0.5**</td>
<td>10.1±0.6**</td>
<td>10.7±0.6**</td>
<td>11.8±0.6**</td>
<td></td>
</tr>
<tr>
<td>+chlorpheniramine</td>
<td>13.8±1.3</td>
<td>9.6±0.5*</td>
<td>9.3±0.6**</td>
<td>9.6±0.6**</td>
<td>11.2±0.9**</td>
<td></td>
</tr>
<tr>
<td>+PC</td>
<td>14.6±1.1</td>
<td>11.1±0.7**</td>
<td>10.9±0.7**</td>
<td>11.4±0.6**</td>
<td>12.4±0.8**</td>
<td></td>
</tr>
<tr>
<td>+CGRP&lt;sub&gt;8–37&lt;/sub&gt;+PC</td>
<td>14.9±1.0</td>
<td>7.3±0.5**</td>
<td>7.3±0.5**</td>
<td>7.4±0.4**</td>
<td>8.1±0.4**</td>
<td></td>
</tr>
</tbody>
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*P<0.05, **P<0.01 vs. antigen, ++P<0.01 vs. preconditioning