0.1 ml S from homogenates tested immediately after 20 min incubation caused significant inhibition of ADP induced aggregation. Adjustment of S after 20 min incubation at pH 12 stabilized inhibitory activity for at least 4 h. This property is shared by pure PGI_2 but the platelet aggregation inhibitory activity of PGD_2 is destroyed at pH 12 in less than 20 min; this would also destroy TxA_2 and PGE_2 activity. By adjusting S to pH 12 for 2 h and neutralizing an estimate of PGI_2-like activity could be obtained without interference from PGD_2 or TxA_2. After 25 min incubation 2 ng PGI_2-like activity/100 mg wet-weight tissue was measured (n =3). The half-life of PGI_2-like inhibitor activity in supernatant was about 10–15 min at 22°C. Maintaining S at room temperature for 1 h or at 37°C for 45 min destroyed 80–90% of the inhibitory activity (n = 20). These properties were also shared by PGI_2 in parallel investigations.

Indomethacin (2 μg/ml) added to brain homogenates had no significant effect on PGI_2-like activity but treatment of rats for 3 days (10 mg kg^{-1} day^{-1}) and addition of 50 μg/ml to incubates abolished PGI_2-like activity. Tranylcypromine (12.5 mg $kg^{-1}day^{-1}$) plus *in vitro* addition of 500 μg/ml failed to modify PGI_2-like activity.

We conclude that a PGI_2-like substance is synthesized in chopped rat brain incubates.

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Prostacyclin-release by bradykinin *in vivo*

S. MONCADA, K.M. MULLANE & J.R. VANE

Department of Prostaglandin Research, Wellcome Research Laboratories, Langley Court, Beckenham, Kent BR3 3BS

Some vascular actions of bradykinin (Bk) have been attributed to prostaglandin generation (see Vane & Ferreira, 1976; McGiff, Terragno, Terragno, Colina & Nasjletti, 1976). The discovery of prostacyclin (PGI_2) and its generation by the vascular wall (Moncada & Vane, 1977) suggests that this substance, rather than PGE_2 is the potential modulator of vascular tone (Dusting, Moncada, Mullane & Vane, 1978). This study was carried out to determine whether Bk releases prostacyclin *in vivo*.

Prostaglandins were detected by the blood-bathed organ technique (Vane, 1964). Blood was continuously withdrawn from catheters placed in the femoral artery or renal vein of chloralose anaesthetized dogs, reoxygenated, and then used to superfuse a bank of assay tissues including bovine coronary artery (BCA), rabbit coeliac (RbCA) or mesenteric artery (RbMA), rabbit aorta (RbA), rat stomach strip (RSS) and rat colon (RC). Propranolol (2 μg/ml), phenoxybenzamine (0.1 μg/ml) and Sar^1-Ile^8)-angiotensin II (0.025 μg/ml) were continuously infused into the blood bathing the tissues to antagonize the effects of catecholamines and angiotensin.

Bk (2–10 μg $kg^{-1}min^{-1}$) infused intravenously or intra-arterially produced dose-related decreases in systemic arterial pressure, the magnitude of the response being independent of the route of administration. The higher doses (5–10 μg $kg^{-1}min^{-1}$) also produced a relaxation of RbCA, RbMA and BCA and contractions of RSS and RbA. Moreover, there was a reduction in the spontaneous activity of RC. Most of the effects on the bioassay tissues could be reproduced by direct infusions of PGI_2 (5–20 ng/ml) into the blood bathing the tissues, while similar infusions of Bk (100–500 ng/ml) contracted the RbA and RSS, and reduced the spontaneity of RC without affecting the other bioassay tissues.

Indomethacin (5 mg/kg, intravenously) abolished the release of the PGI_2-like substance but did not modify the peak hypotensive response to intravenous Bk, but reduced the sustained fall in blood pressure. In contrast, inhibition of converting enzyme with SQ14,225 (0.1–0.2 mg/kg) led to an enhanced release of a PGI_2-like substance to subsequent infusions of Bk (0.1–0.5 μg $kg^{-1}min^{-1}$) (3 experiments).

When infused into the kidney Bk (1–5 μg $kg^{-1}min^{-1}$) produced an increase in renal blood flow concomitant with the appearance in renal venous blood of a substance with the same bioassay profile as PGI_2 (2 experiments). In one experiment, acute nephrectomy prevented prostacyclin release due to intravenous bradykinin.

These results suggest that bradykinin selectively increases PGI_2 release; the kidney is the main organ involved, and finally that the released prostacyclin contributes to the sustained hypotension produced by intravenous Bk in the dog.

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Activity of prostacyclin, a stable analogue, 6ß-PGI_1 and 6-oxo-$PGF_{1\alpha}$ on canine isolated parietal cells

A.H. SOLL & B.J.R. WHITTLE

Wadsworth V.A. Hospital, U.C.L.A. School of Medicine, Los Angeles, U.S.A.
and
Department of Prostaglandin Research, Wellcome Research Laboratories, Langley Court, Beckenham Kent BR3 3BS

Prostacyclin (PGI_2) and its stable 5–6 dihydro analogue, 6β-PGI_1, inhibit acid secretion from the perfused gastric mucosa of the rat *in vivo* and *in vitro* (Whittle, Boughton-Smith, Moncada & Vane, 1978). The present study concerns the activity of prostacyclin, its chemical breakdown product 6-oxo-$PGF_{1\alpha}$ and 6β-PGI_1 on [^{14}C]-aminopyrine accumulation, an index of acid secretory activity (Berglindh, Helander & Obrink, 1976; Soll, 1978a) in canine isolated parietal cells. Further, since the anti-secretory action of prostaglandin E_2 has previously been linked to reduction in the parietal cell level of cyclic AMP, (cAMP) the potential intracellular mediator of gastric secretion (Soll, 1978a; Major & Scholes, 1978), the effects of prostacyclin on cAMP levels and concomitant parietal-cell function were investigated.

Cells were isolated from the acid-secreting fundic area of the canine mucosa by sequential treatment with crude collagenase and EDTA, and a parietal-cell enriched (60–80% of the total cells) fraction was obtained from an elutriator rotor using a sedimentation velocity technique (Soll, 1978b). Cells were incubated for 20 min in a modified Hanks' medium (pH 7.4 at 37°C) containing histamine, and freshly prepared prostacyclin or the other prostaglandins. [^{14}C]-Aminopyrine ([^{14}C]-AP) accumulation, which depends on the ionization and trapping of this weak base within the parietal-cell during hydrogen-ion elaboration and secretion was measured in aliquots of the cells, and cAMP was determined by radio-immunoassay (Soll, 1978a).

Histamine (10^{-7} – 10^{-4} M) caused a dose-dependent uptake and trapping of [^{14}C]-AP in the cells, as previously shown (Soll, 1978a) and this was inhibited in a dose-dependent manner by concurrent incubation with the prostaglandin (10^{-9} – 10^{-5} M). Using a sub-maximal dose of histamine (10^{-5} M), the ID_{50} (dose causing 50% inhibition) was 0.9×10^{-6} M for prostacyclin, 5×10^{-7} M for 6β-PGI_1, 5×10^{-4} M for 6-oxo-$PGF_{1\alpha}$ and 10^{-8} M for PGE_2. The lower activity of prostacyclin compared to PGE_2 could reflect the rapid breakdown of prostacyclin under the incubation conditions used. In other systems where stability is of limited importance, prostacyclin is some 500–1,000 times more active than 6-oxo-$PGF_{1\alpha}$ and 20–200 times more active than 6β-PGI_1 (Whittle, *et al.*, 1978) whereas in the present study prostacyclin was less potent than this stable analogue.

Histamine (10^{-5} M) increased the cAMP levels in the parietal-cell fraction and this was dose-dependently inhibited by simultaneous incubation with prostacyclin (10^{-8} – 10^{-5} M) in the same concentration range which reduced the parietal-cell accumulation of ^{14}C-AP. As with PGE_2, higher concentrations (10^{-4} M) of prostacyclin revealed a biphasic relationship, with cAMP concentrations becoming elevated. This could reflect stimulation of adenylate cyclase in the few contaminating cells or the parietal cells themselves at high prostaglandin concentrations.

The present observations suggest that the potent gastric antisecretory actions of prostacyclin and its analogue *in vivo* (Whittle, *et al.*, 1978) are the result of direct effects on the parietal cells. As previously suggested for PGE_2 (Soll, 1978a; Major & Scholes, 1978) prostacyclin may exert its gastric antisecretory actions by a direct inhibitory effect on parietal-cell adenylate cyclase. It is not yet known whether prostacyclin, which can be generated by the gastric mucosa (Moncada, Salmon, Vane & Whittle, 1978), can act as an endogenous modulator of parietal cell function *in vivo*.

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