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Enhancement by endothelin-1 of microvascular permeability via the activation of ET_A receptors

[1]János G. Filep, Martin G. Sirois, Éva Földes-Filep, Anne Rousseau, Gérard E. Plante, *Alain Fournier, †Mitsuo Yano & Pierre Sirois

Department of Pharmacology, Faculty of Medicine, University of Sherbrooke, Sherbrooke, P.Q., Canada J1H 5N4; *Institut National de la Recherche Scientifique-Santé, Montréal, P.Q., Canada H9R 1G6 and †Central Research Laboratories, Banyu Pharmaceuticals Co., Tsukuba, Japan

1 The objective of the present experiments was to assess the involvement of endothelin-A (ET_A) receptors in mediating the effects of endothelin-1 on microvascular permeability in conscious rats.

2 Bolus injection of endothelin-1 (0.1 and 1 nmol kg^{-1}, i.v.) resulted in a dose-dependent prolonged pressor effect preceded by a transient depressor response. These changes were accompanied by a dose-dependent loss of plasma volume. Endothelin-1 (1 nmol kg^{-1}) enhanced the vascular permeability of the upper and lower bronchi, kidney, stomach, duodenum and spleen (up to 270%) as measured by the extravasation of Evans blue dye.

3 Pretreatment of the animals with the selective ET_A receptor antagonist, BQ-123 (1 mg kg^{-1}, i.v.) significantly blunted the pressor response to endothelin-1 without affecting the depressor response. BQ-123 inhibited by 87% the endothelin-1 (1 nmol kg^{-1})-induced plasma volume loss. BQ-123 markedly attenuated protein extravasation elicited by endothelin-1 in the upper and lower bronchi and kidney, whereas it completely inhibited the permeability effect of endothelin-1 in the stomach and duodenum. BQ-123 by itself had no significant effect on the parameters studied.

4 The endothelin-1 analogue, [Trp(For)^{21}]-endothelin-1, in which Trp^{21} is formylated, was as potent a pressor agent as endothelin-1, but had no depressor action. Bolus injection of [Trp(For)^{21}]-endothelin-1 (0.1 and 1 nmol kg^{-1}, i.v.) evoked similar plasma volume losses to those observed following administration of equimolar doses of endothelin-1. Furthermore, 1 nmol kg^{-1} [Trp(For)^{21}]-endothelin-1 evoked increases in protein extravasation similar to endothelin-1, 1 nmol kg^{-1}.

5 The present findings suggest that endothelin-1 enhances microvascular permeability, in part, via the activation of ET_A receptors.

Keywords: Endothelin-1; ET_A receptor; protein extravasation; haemoconcentration; airways; gastrointestinal tract; kidney; BQ-123

Introduction

An increasing body of evidence suggests that, in addition to modulation of vascular tone, endothelin-1 may also play a role in the regulation of vascular permeability. Endothelin-1 causes an increase in haematocrit not fully accounted for by urinary fluid loss (Goetz *et al.*, 1988; Miller *et al.*, 1989; López-Farré *et al.*, 1989), suggesting that this haemoconcentration could result from vascular leakage. Subsequent studies have shown endothelin-1-induced oedema formation in the forearm in man (Dahlöf *et al.*, 1990) and enhanced protein extravasation in selected vascular beds including the airways, heart, gastrointestinal tract and kidney in the rat (Filep *et al.*, 1991; 1992a; Zimmerman *et al.*, 1992). In other studies, endothelin-1 was found to attenuate oedema formation elicited by chemotactic agents in rat and rabbit skin (Brain *et al.*, 1989; Chander *et al.*, 1989) and in the mouse paw (Henriques *et al.*, 1992).

Kinetic analysis of endothelin-1 binding to various tissues, functional data and cross-desensitization experiments implied the possible existence of multiple endothelin receptor subtypes (Sakurai *et al.*, 1992; Sokolovsky, 1992). Recently, the existence of at least two distinct types of endothelin receptors has been demonstrated by cloning cDNAs encoding those subtypes (Arai *et al.*, 1990; Sakurai *et al.*, 1990). The ET_A receptor is highly selective for endothelin-1 (Arai *et al.*, 1990), whereas the ET_B receptor is non-isopeptide-selective (Sakurai *et al.*, 1990). Endothelin receptors mediating vasoconstriction are thought to be of the ET_A receptor subtype; ET_B receptors have been suggested to mediate the depressor and vasodilator responses to endothelin-1 (Ihara *et al.*, 1991; Sakurai *et al.*, 1992). No information is available on the receptors mediating the permeability effect of endothelin-1.

The present study was attempted to identify the receptors responsible for the microvascular permeability effects of endothelin-1 in the conscious rat. In these experiments we have studied the effects of the selective ET_A receptor antagonist, BQ-123 (Ihara *et al.*, 1992) on protein extravasation elicited by endothelin-1 in selected vascular beds and compared the permeability effects of endothelin-1 and its analogue, [Trp(For)^{21}]-endothelin-1, which possesses the pressor but not the depressor activity of endothelin-1 (Filep *et al.*, 1992b).

Methods

The experiments were performed on conscious, chronically catheterized male Wistar rats weighing 225–290 g. The animals were kept in individual metabolic cages and were prepared as described previously (Filep *et al.*, 1987). Briefly, under anaesthesia (ketamine, 75 mg kg^{-1} and sodium pentobarbitone, 15 mg kg^{-1}) catheters were implanted into the abdominal aorta and vena cava through the central tail artery and left femoral vein, respectively. The venous catheter

[1] Author for correspondence at present address: Research Center, Maisonneuve-Rosemont Hospital, University of Montréal, 5415 boul. de l'Assomption, Montréal, P.Q., Canada H1T 2M4.

was led subcutaneously to the root of the tail. The catheters emerging from the tail were protected by an acrylic cuff glued to the tail. The cuff was connected to a stainless steel spiral and was fed through the top of the metabolic cage. The animals were allowed to recover completely for at least 4 days following the surgical procedures. During the experiments the animals could move freely and had free access to food and water. Mean arterial blood pressure (MABP) was monitored continuously with a Statham P23 dB pressure transducer. Protein extravasation was estimated by measuring the extravasation of Evans blue dye, which binds to plasma albumin (Rawson, 1943).

Experimental protocols

On the day of the experiment, following an equilibrium period of 1 h, basal cardiovascular parameters were measured for 20 min before drug administration. To measure protein extravasation, Evans blue dye (20 mg kg^{-1}, 25 mg ml^{-1} in 0.9% NaCl) was injected i.v. together with either endothelin-1 (1 nmol kg^{-1}), the endothelin-1 analogue, [Trp(For)^{21}] endothelin-1, in which Trp^{21} was formylated (1 nmol kg^{-1}) or their vehicle. Previous experiments showed that at a dose of 1 nmol kg^{-1}, endothelin-1 enhances protein extravasation in the rat pulmonary, renal and mesenteric circulation (Filep *et al.*, 1991). The endothelin-1 analogue, [Trp(For)^{21}]endothelin-1 is as potent a pressor agent as the mother compound, but it is devoid of the depressor activity in conscious rats (Filep *et al.*, 1992b). Some animals were pretreated with 1 mg kg^{-1} of the selective ET_A receptor antagonist, BQ-123 (Ihara *et al.*, 1992), for 5 min before injection of endothelin-1. Twenty min before, and 10 min after, injection of endothelin-1, blood (approximately 15 µl) was collected through the arterial catheter into heparinized glass capillaries to determine the haematocrit. Ten min after injection of Evans blue dye, the animals were anaesthetized (sodium pentobarbitone, 50 mg kg^{-1}) and were perfused with 40 ml 0.9% NaCl through a catheter inserted into the abdominal aorta. Then the thorax was cut open, the trachea, upper bronchi (airways extending from the bifurcation of trachea to its entry into the parenchyma), lower bronchi (defined as major airways surrounded by parenchyma that can be easily dissected without magnification) and portions of pulmonary parenchyma were prepared. Portions of liver, spleen, pancreas, kidney, stomach and duodenum were excised and weighed. Tissue Evans blue dye content was measured by spectrophotometry following extraction with formamide (4 ml per g wet tissue weight) as described previously (Filep *et al.*, 1991). The Evans blue content of each sample was expressed as µg dye per g dry weight of tissue to avoid underestimation of changes due to plasma fluid extravasation.

Drugs and chemicals

Endothelin-1 and [Trp(For)^{21}]endothelin-1 were synthesized in our laboratories by solid phase methodology. The purity of the preparations were greater than 97% as measured by high performance liquid chromatography. Endothelin-1 and its analogue were dissolved in distilled water and stored under N_2 at − 20°C for up to 4 weeks. On the day of the experiments, an aliquot was removed and diluted further with 0.9% NaCl. BQ-123(-cyclo-(D-Asp-L-Pro-D-Val-L-Leu-D-Trp-)sodium (Banyu Pharmaceuticals Co., Tsukuba, Japan) was dissolved in 0.9% NaCl immediately before use. Evans blue dye was from Sigma Chemical Co. (St Louis, MO, U.S.A.).

Data analysis

Results are expressed as means ± s.e.mean. Changes in plasma volume were calculated according to the formula: Δplasma volume (%) = (100/100-Hct_i) × ((Hct_i-Hct_f)/Hct_f) × 100, where Hct_i and Hct_f are the haematocrit values obtained 20 min before (initial or control) and 10 min after (final Hct) injection of endothelin-1, respectively.

Statistical evaluation of the data was performed by two-way analysis of variance using ranks (Friedman's test) followed by a Wilcoxon-Wilcox test (Wilcoxon & Wilcox, 1964), to identify differences between control and repeated measurements on the same animals; by Dunn's multiple contrast hypothesis test (Dunn, 1964), when various treatments were compared to the same control; or by Wilcoxon signed rank test and Mann-Whitney U test, for paired and unpaired observations, respectively. A level of $P<0.05$ was considered significant for all tests.

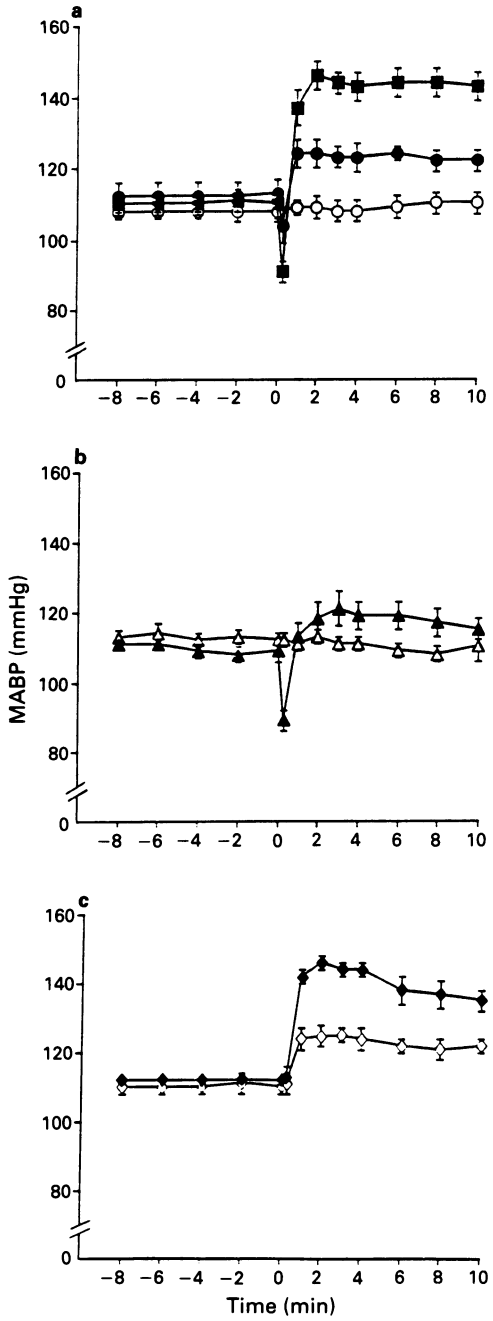

Figure 1 Blood pressure responses to endothelin-1 and [Trp(For)^{21}]-endothelin-1 in the absence and presence of BQ-123 in the conscious rat. Various doses of endothelin-1 or its analogue were injected i.v. at 0 min and BQ-123 was injected at − 5 min as follows: (a) 0.9% NaCl (○) ($n = 6$); endothelin-1, 0.1 nmol kg^{-1} (●) ($n = 5$) or 1 nmol kg^{-1} (■) ($n = 6$); (b) BQ-123, 1 mg kg^{-1} plus 0.9% NaCl (△) ($n = 5$); BQ-123, 1 mg kg^{-1} plus endothelin-1, 1 nmol kg^{-1} (▲) ($n = 6$) and (c) [Trp(For)^{21}]-endothelin-1, 0.1 nmol kg^{-1} (◇) ($n = 5$) or 1 nmol kg^{-1} (◆) ($n = 5$). Values are means ± s.e.mean.

Results

Effects on arterial blood pressure and plasma volume

Bolus i.v. injections of endothelin-1 (0.1 and 1 nmol kg^{-1}) evoked prolonged dose-dependent increase in MABP preceded by a transient hypotension (Figure 1a). Administration of BQ-123 (1 mg kg^{-1}) by itself did not produce significant changes in MABP, whereas it markedly inhibited the pressor action of endothelin-1 (maximum increases in MABP were 37 ± 1 and 11 ± 3 mmHg in the absence and presence of BQ-123 following 1 nmol kg^{-1} endothelin-1, respectively, $n = 6$, $P < 0.01$) without affecting the depressor response to endothelin-1 (Figure 1b). Increasing the dose of BQ-123 did not cause further inhibition of the pressor effect of endothelin-1. The endothelin-1 analogue, [Trp(For)^{21}]-endothelin-1 appeared to be as effective as a pressor agent as the parent peptide, but, unlike endothelin-1, it did not induce a depressor response (Figure 1c).

Endothelin-1 and [Trp(For)^{21}]-endothelin-1-induced changes in MABP were accompanied by a dose-dependent haemoconcentration. At the dose of 0.1 nmol kg^{-1}, neither endothelin-1 nor [Trp(For)^{21}]-endothelin-1 affected the haematocrit significantly. Following administration of 1 nmol kg^{-1} endothelin-1 or its analogues, the haematocrit increased on average by 11 and 8%, corresponding to about 17 and 12% plasma volume loss, respectively (Table 1). Pretreatment of the animals with BQ-123, 1 mg kg^{-1} resulted in 87% reduction in endothelin-1-induced plasma volume loss. BQ-123 by itself had no significant effect on haematocrit (Table 1).

Effects on protein extravasation

To study the effects of BQ-123 on endothelin-1-induced protein extravasation, the animals received 1 mg kg^{-1} BQ-123 5 min before the combined injection of endothelin-1, 1 nmol kg^{-1} and Evans blue dye (20 mg kg^{-1}). BQ-123 reduced the extravasation of Evans blue dye by 42, 66, 100, 100 and 79% in the upper and lower bronchi, stomach, duodenum and kidney, respectively (Figures 2 and 3). Although the mean increase in protein extravasation in the spleen was lower after pretreatment with BQ-123, the effect of endothelin-1 on tissue Evans blue content did not differ significantly from that seen after endothelin-1 alone (Figure 3). BQ-123 by itself did not affect extravasation of Evans blue in the vascular beds studied (Figures 2 and 3).

Injection of 1 nmol kg^{-1} [Trp(For)^{21}]-endothelin-1 increased tissue Evans blue content in the upper and lower bronchi on average by 80 and 133%, respectively (Figure 2). Although Evans blue content tended to be higher in the trachea and pulmonary parenchyma, these changes did not reach statistical significance (Figure 2). [Trp(For)^{21}]-endothelin-1 injection produced increases of 68, 102, 105, 62 and 125% in Evans blue content in the liver, stomach, duodenum, spleen and kidney, respectively (Figure 2). The endothelin-1 analogue induced changes in protein extravasation did not differ significantly from those observed following endothelin-1 (Figures 2 and 3). Like endothelin-1, [Trp(For)^{21}]-endothelin-1 did not enhance protein extravasation in the pancreas, skeletal muscle and skin (Figures 3 and 4).

Discussion

The results of the present study confirm that the effects of intravenously injected endothelin-1 increase haematocrit (Goetz *et al.*, 1988; Miller *et al.*, 1989, Filep *et al.*, 1991) and protein extravasation in various organs (Filep *et al.*, 1991; Zimmerman *et al.*, 1992) and provide evidence for the involvement of ET_A receptors in mediating the permeability effect of endothelin-1.

Bolus injection of endothelin-1 i.v. resulted in haemoconcentration as evidenced by an increase of about 10% in haematocrit, reflecting a reduction of about 17% in plasma volume. This plasma volume loss was almost completely prevented by pretreatment of the animals with BQ-123. BQ-123 has been reported to be 2500 times more potent in inhibiting the binding of endothelin-1 to ET_A than ET_B receptors *in vitro* (Ihara *et al.*, 1992). Furthermore, BQ-123 significantly attenuated the pressor response to endothelin-1 without affecting its depressor action (Ihara *et al.*, 1992 and the present study). Since ET_B receptors located on the vascular endothelium have been implicated in the mediation of the transient depressor action of endothelin-1 (Saeki *et al.*, 1991; Douglas & Hiley, 1991a), it might be assumed that BQ-123 does not interfere with ET_B receptors *in vivo*. It should be noted, however, that the degree of inhibition of the pressor effect of endothelin-1 by BQ-123 did not exceed 70%. A previous study showed that increasing the dose of BQ-123 (up to 10 mg kg^{-1}) did not reduce further the maximum increase in MABP elicited by endothelin-1, rather it shortened the duration of the pressor response to endothelin-1 (Ihara *et al.*, 1992). Furthermore, [Ala^{1,3,11,15}]endothelin-1, a selective ET_B receptor agonist (Saeki *et al.*, 1991) has been reported to evoke pressor responses in anaesthetized rats (Douglas & Hiley, 1991a). These observations lend further support to the notion that more than one type of endothelin receptor is involved in the generation of the pressor effect, as

Table 1 Endothelin-1-induced haemoconcentration and its inhibition by BQ-123 in the conscious rat

	n	*Haematocrit (vol %)* *Before endothelin-1*	*10 min after injection*	P^a	*Δ Plasma volume (%)*	P^b
Vehicle	6	40.5 ± 0.5	40.6 ± 0.3	NS	− 0.6 ± 1.4	–
Endothelin-1 (0.1 nmol kg^{-1})	5	39.9 ± 0.6	40.4 ± 0.4	NS	− 2.1 ± 2.0	NS
Endothelin-1 (1 nmol kg^{-1})	6	40.1 ± 0.6	44.6 ± 0.5	0.05	− 16.6 ± 2.8	0.05
BQ-123 (1 mg kg^{-1})	5	41.1 ± 0.9	41.1 ± 0.9	NS	− 0.1 ± 2.7	NS
BQ-123 (1 mg kg^{-1}) + endothelin-1 (1 nmol kg^{-1})	6	40.8 ± 1.1	41.6 ± 0.6	NS	− 2.2 ± 3.2	NS
[Trp(For)^{21}]-endothelin-1 (0.1 nmol kg^{-1})	5	41.6 ± 0.5	42.2 ± 0.7	NS	− 2.6 ± 2.4	NS
[Trp(For)^{21}]-endothelin-1 (1 nmol kg^{-1})	5	41.6 ± 0.9	44.8 ± 0.5	0.05	− 12.1 ± 2.4	0.05

Values are means ± s.e.mean

[a]*P* values were obtained by the Wilcoxon's signed rank test; [b]compared to vehicle by the Dunn's multiple contrast hypothesis test.

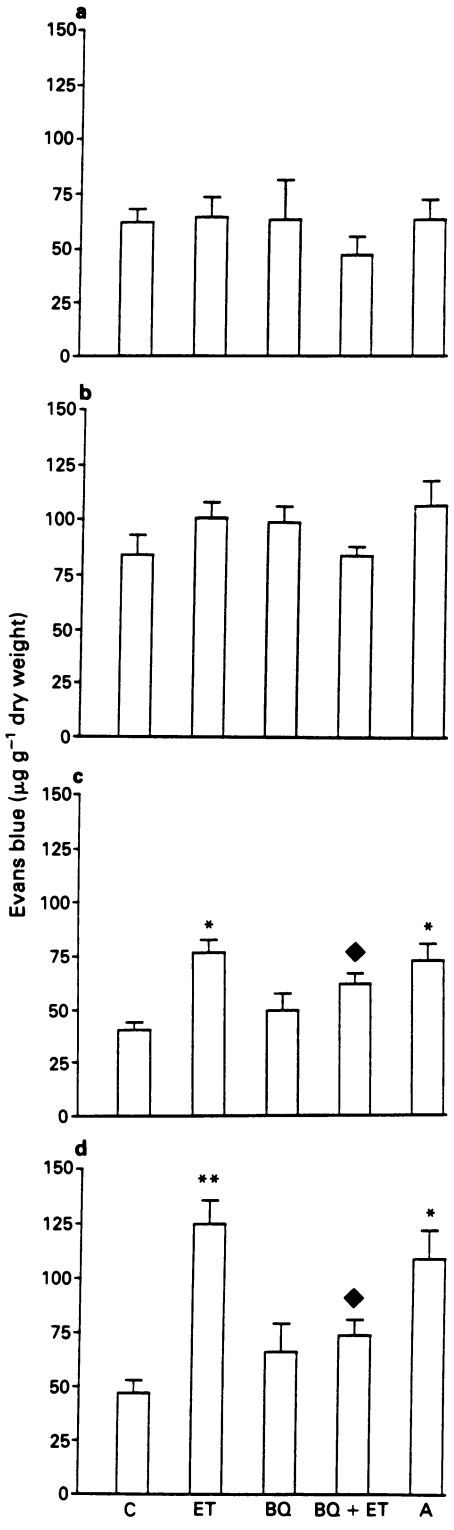

Figure 2 Effects of endothelin-1, BQ-123 and [Trp(For)^{21}]-endothelin-1 on protein extravasation in rat airways: (a) trachea, (b) parenchyma, (c) upper bronchi and (d) lower bronchi. The animals were injected i.v. with 0.9% NaCl (C), endothelin-1 (ET, 1 nmol kg^{-1}), BQ-123 (BQ, 1 mg kg^{-1}), BQ-123 (1 mg kg^{-1}, 5 min prior to endothelin-1) plus endothelin-1, (1 nmol kg^{-1}) or [Trp(For)^{21}]-endothelin-1 (A, 1 nmol kg^{-1}) together with Evans blue dye (20 mg kg^{-1}). The rats were killed 10 min after injection of the dye. Values are means with s.e.mean. $n = 5$ for BQ-123 and [Trp(For)^{21}]-endothelin-1, $n = 6$ for the other groups. $^*P < 0.05$; $^{**}P < 0.01$ (compared to control); ◆$P < 0.05$ (compared to endothelin-1).

has been demonstrated for endothelin-1-induced vasoconstriction in the pig coronary artery (Harrison *et al.*, 1992).

Although immunoreactive endothelin-1 can be detected in normal rat plasma (Shirakami *et al.*, 1991; Valentin *et al.*, 1991), little is known about the physiological importance of circulating endothelin-1. Administration of BQ-123 neither produced changes in MABP nor affected haematocrit and protein extravasation. Thus, it seems unlikely that circulating endothelin-1 could support blood pressure, or modulate microvascular permeability through the ET_A receptor under physiological conditions, or both. On the other hand, BQ-123 significantly attenuated the accumulation of Evans blue dye, which is bound by plasma albumin, in various organs after injection of endothelin-1. However, BQ-123 appeared to be a more effective inhibitor in the stomach, duodenum and kidney than in the upper and lower bronchi and spleen. In the stomach and duodenum, BQ-123 completely inhibited endothelin-1-induced protein extravasation, whereas only 40% inhibition was observed in the spleen. These findings suggest that different endothelin receptors or mechanisms might mediate the permeability effect of endothelin-1 in different vascular beds. Indeed, endothelin-3 has been reported to promote extravasation of Evans blue dye in skeletal muscle (Valentin *et al.*, 1991), where endothelin-1 failed to alter vascular permeability. It is also possible that the bioavailability of BQ-123 might be different in various organs.

Additional evidence supporting the involvement of ET_A receptors in mediating the permeability effect of endothelin-1 is derived from the experiments with [Trp(For)^{21}]-endothelin-1. We have previously reported that formylation of the terminal Trp in endothelin-1 leads to an analogue which retains the pressor activity of endothelin-1, but is devoid of the depressor action (Filep *et al.*, 1992b). The lack of depressor activity may suggest that [Trp(For)^{21}]-endothelin-1 does not act on ET_B receptors. In addition to being as potent a pressor agent as the parent peptide, [Trp(For)^{21}]-endothelin-1 evoked similar increases in haematocrit as equimolar doses of endothelin-1. Furthermore, [Trp(For)^{21}]-endothelin-1 mimicked the effect of endothelin-1 on accumulation of Evans blue dye in the upper and lower bronchi, stomach, duodenum, kidney and spleen, whereas it appeared somewhat more effective than endothelin-1 in the liver. The reason for this latter finding is not clear. However, no other difference in the actions of endothelin-1 and its analogue was observed in the present study. Thus, like endothelin-1, [Trp(For)^{21}]-endothelin-1 did not affect microvascular permeability in the trachea, pulmonary parenchyma, pancreas, skin and skeletal muscle.

In the present study parallel changes were observed in vascular permeability and MABP. Thus, enhanced protein extravasation was detected when MABP was elevated following administration of endothelin-1 or [Trp(For)^{21}]-endothelin-1, and protein extravasation was inhibited when the pressor response to endothelin-1 was attenuated. However, the increase in blood pressure, *per se*, could not be the basis for the observed protein extravasation. Endothelin-1 was found to be a considerably more potent constrictor of venous than arterial vessels (Yang *et al.*, 1989) and it has a longer duration of action in the venous than arterial vasculature (Warner, 1990). An increase in both pre-, and post-capillary resistance could lead to an increase in capillary hydrostatic pressure. This might explain an enhanced transcapillary fluid transfer, but not an increased protein leakage. Mediator-stimulated increase in protein extravasation is primarily attributable to an increase in the hydraulic conductivity of the microvascular membrane secondary to interendothelial cell gap formation in the venules (Grega *et al.*, 1986). Vasoactive mediators which can elevate microvascular pressure, but do not elicit gap formation could not promote protein efflux and oedema formation (Grega *et al.*, 1986). Accordingly, it has been suggested that endothelin-1 induces gap formation directly or through release of secondary mediators such as platelet-activating factor (Filep *et al.*, 1991) or thromboxane

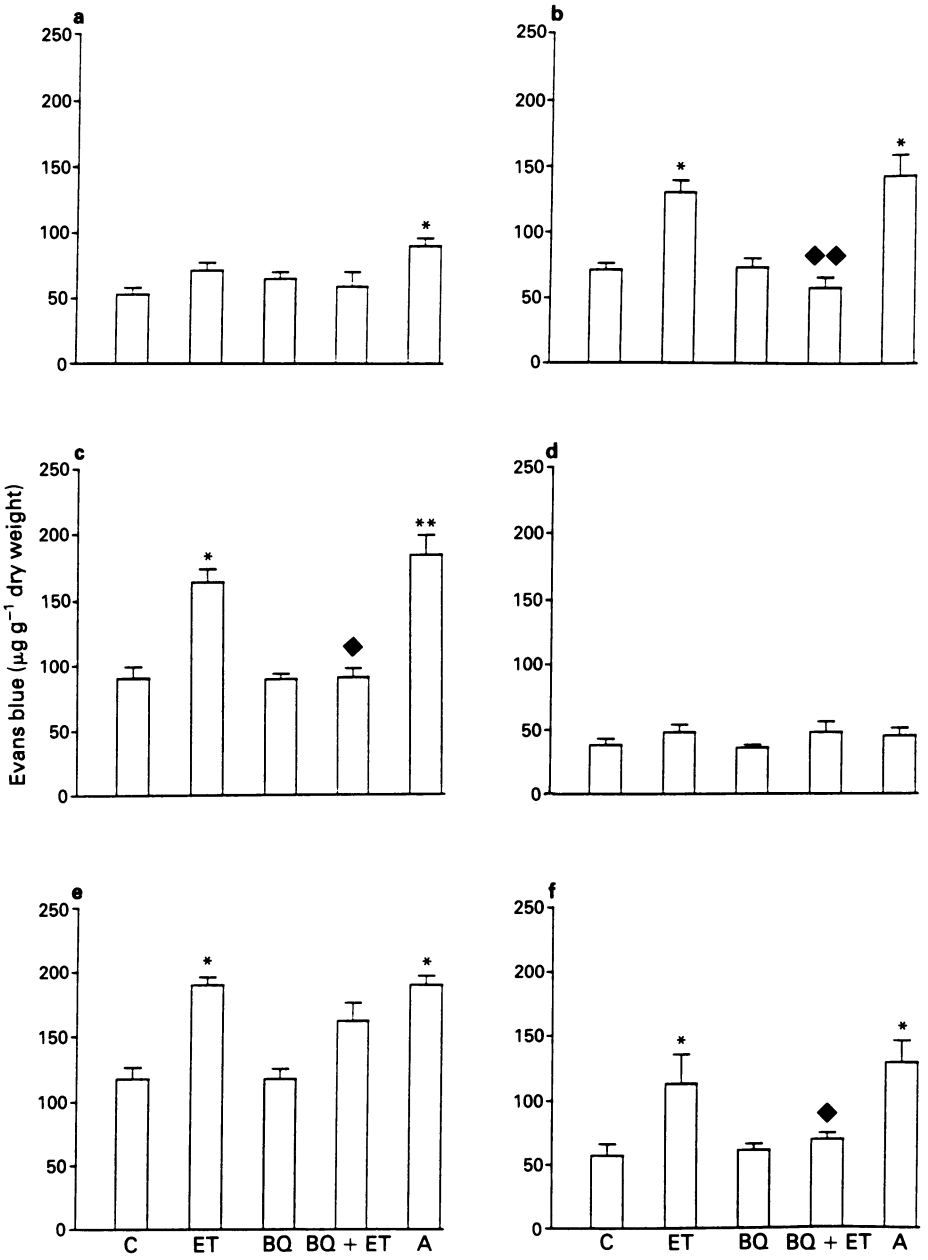

Figure 3 Effects of endothelin-1, BQ-123 and [Trp(For)^{21}]-endothelin-1 on protein extravasation in the rat liver (a), stomach (b), duodenum (c), pancreas (d), spleen (e) and kidney (f). The animals were given i.v. 0.9% NaCl (C), endothelin-1 (ET, 1 nmol kg^{-1}), BQ-123 (BQ, 1 mg kg^{-1}), BQ-123 (1 mg kg^{-1}, 5 min prior to endothelin-1) plus endothelin-1 (1 nmol kg^{-1}) or [Trp(For)^{21}]-endothelin-1 (A, 1 nmol kg^{-1}) together with Evans blue dye (20 mg kg^{-1}). The rats were killed 10 min after injection of the dye. Values are means with s.e.mean. $n = 5$ for BQ-123 and [Trp(For)^{21}]-endothelin-1, $n = 6$ for the other groups. *$P<0.05$; **$P<0.01$ (compared to control); ◆$P<0.05$; ◆◆$P<0.01$ (compared to endothelin-1).

A_2 (Sirois *et al.*, 1992). Changes in capillary pressure are not primary determinants of macromolecular permeability (Amelang *et al.*, 1981; Grega *et al.*, 1986). Indeed, endothelin-1-induced protein extravasation was markedly attenuated by receptor antagonists to platelet-activating factor and thromboxane A_2, which did not alter blood pressure responses to endothelin-1 (Filep *et al.*, 1991; Sirois *et al.*, 1992). Moreover, an increase in haematocrit has also been observed after the infusion of a non-pressor dose of endothelin-1 (López-Farré *et al.*, 1989). Although the permeability effect of endothelin-1 appears to be independent of changes in arterial blood pressure, dramatic increases in microvascular pressure might lead to disruption of endothelial junctions, resulting in extravasation of erythrocytes as it has been observed following intra-arterial injections of large doses of endothelin-1 into the rat mesenteric vascular bed (Douglas & Hiley, 1991b).

The potent arteriolar constrictor effect of endothelin-1 could mask any oedema-inducing effect of the peptide as has been suggested for the skin (Brain *et al.*, 1988). Furthermore, topical application of endothelin-1 reduced oedema formation elicited by chemotactic agents in the rat and rabbit skin (Brain *et al.*, 1989; Chander *et al.*, 1989). However, ET_A receptor blockade with BQ-123 did not lead to an increase in protein accumulation in the skin. These findings suggest that either the constrictor action of endothelin-1 is not mediated through ET_A receptors in the skin or endothelin-1 does not

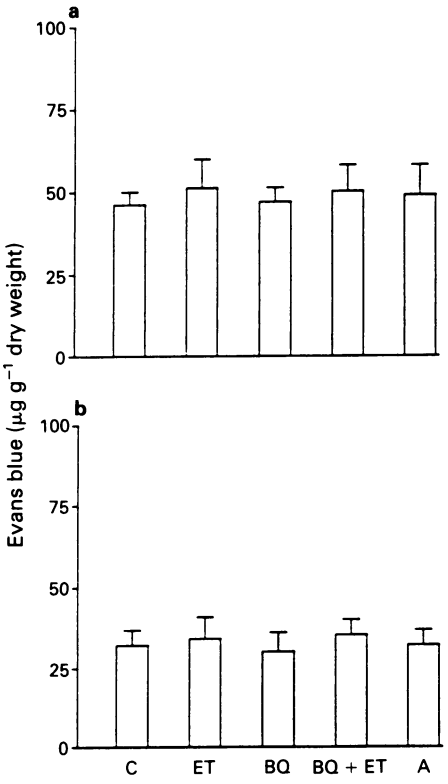

Figure 4 Evans blue content in the rat skeletal muscle (right quadriceps) (a) and dorsal skin (b) following endothelin-1, BQ-123 and [Trp(For)^{21}]-endothelin-1. The animals were given i.v. 0.9% NaCl (C), endothelin-1 (ET, 1 nmol kg^{-1}), BQ-123 (BQ, 1 mg kg^{-1}), BQ-123 plus endothelin-1 or [Trp(For)^{21}]-endothelin-1 (A, 1 nmol kg^{-1}) together with Evans blue dye. The rats were killed 10 min after the injection of the dye. Values are means with s.e.mean. $n = 5$ for BQ-123 and [Trp(For)^{21}]-endothelin-1, $n = 6$ for the other groups.

enhance permeability in this tissue. The different susceptibility of different vascular beds to endothelin-1 and the differences in the inhibitory effect of BQ-123 on protein accumulation evoked by endothelin-1 may reflect differential sensitivity of the vasculature to endothelin-1 or differences in distribution of endothelin receptor subtypes.

In conclusion, the present data show that endothelin-1-induced haemoconcentration and increase in protein extravasation is mediated, in part, through activation of the ET_A receptor. The different susceptibility of various vascular beds to the inhibitory action of BQ-123, however, suggest that in certain vascular beds this might not be the sole mechanism by which endothelin-1 enhances vascular permeability.

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