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Effects of inhibitors of arachidonic acid metabolism on Paf-induced gastric mucosal necrosis and haemoconcentration

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1 The effects of several inhibitors of arachidonic acid metabolism on gastric necrosis, hypotension, haemoconcentration, leukopenia and plasma exudation induced by platelet-activating factor (Paf) were studied in the rat.

2 A 10 min intravenous infusion of Paf (100 ng kg^{-1} min^{-1}) caused extensive gastric damage and a marked fall in systemic blood pressure which had not recovered to basal levels 30 min after the infusion had been terminated. Paf also caused significant haemoconcentration, plasma exudation and transient leukopenia.

3 Pretreatment with dexamethasone (0.2 or 2 mg kg^{-1} s.c.) or prednisolone (20 mg kg^{-1} s.c.) two hours before Paf significantly reduced the gastric damage and accelerated the recovery of blood pressure after the Paf infusion. Likewise, BW755C (50 mg kg^{-1} p.o.) significantly reduced the gastric damage.

4 Acute pretreatment with dexamethasone (2 mg kg^{-1} i.v.) 15 min before Paf, or with indomethacin (5 mg kg^{-1} s.c.), acetylsalicylic acid (10 mg kg^{-1} i.v.) or l-benzylimidazole (50 mg kg^{-1} s.c.) did not significantly affect the gastric damage induced by Paf.

5 The Paf-induced haemoconcentration and plasma exudation were significantly reduced by pretreatment with prednisolone (20 mg kg^{-1} s.c.) or BW755C (50 mg kg^{-1} p.o.), while Paf-induced leukopenia was unaffected by either drug.

6 These studies indicate that cyclo-oxygenase products of arachidonic acid are unlikely to contribute significantly to the gastric damage or the prolonged hypotension induced by Paf. The ability of corticosteroids and BW755C to reduce the gastric damage, haemoconcentration and plasma exudation suggests that lipoxygenase products of arachidonic acid may contribute to these actions of Paf.

Introduction

Platelet-activating factor (Paf) is a putative mediator of inflammation and anaphylaxis (Benveniste, 1974; Vargaftig *et al.*, 1981). Recently, we demonstrated that this low molecular weight phospholipid was a potent ulcerogenic agent when infused intravenously to rats (Rosam *et al.*, 1986). Furthermore, doses of Paf as low as 2 pmol kg^{-1} significantly augmented the extent of gastric damage induced by topically applied 20% ethanol (Wallace & Whittle, 1986a). The mechanism of Paf-induced gastric mucosal necrosis is not yet clear. However, a mechanism involving effects of Paf on platelets is unlikely since Paf does not bind to rat platelets (Inarrea *et al.*, 1985) and because Paf causes gastric damage in thrombocytopenic rats (Rosam *et al.*, 1986).

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The common histological feature of Paf-induced gastric and intestinal damage is congestion of mucosal microvessels (Rosam *et al.*, 1986; Wallace & Whittle, 1986b). It is possible that this vasocongestion occurs as a result of the ability of Paf to increase vascular permeability (Bessin *et al.*, 1983; Doebber *et al.*, 1984). These changes in vascular permeability may be mediated or amplified by release of eicosanoids, such as the leukotrienes (Dahlen *et al.*, 1981) or vasodilator prostaglandins (Archer *et al.*, 1984; McGivern *et al.*, 1984). In addition, prostaglandins of the E and I series exert protective actions in the gastric mucosa (Robert *et al.*, 1979) while other products of the arachidonic acid cascade (thromboxane A_2, leukotriene C_4) are pro-ulcerogenic (Whittle *et al.*, 1981; 1985). We have therefore examined the possible contribution of arachidonic acid metabolites to Paf-induced gastric

necrosis. To study this role, several inhibitors of arachidonic acid metabolism have been utilized, including corticosteroids (dexamethasone and prednisolone), cyclo-oxygenase inhibitors (indomethacin and acetylsalicyclic acid), a thromboxane synthase inhibitor (1-benzylimidazole) and a dual cyclo-oxygenase/lipoxygenase inhibitor (BW755C). In addition, the effects of some of these drugs on Paf-induced leukopenia, haemoconcentration and plasma exudation were studied.

Methods

Male, Wistar rats (220–250 g) were deprived of food, but not water, for 18–20 h prior to an experiment. The rats were anaesthetized with sodium pentobarbitone (60 mg kg^{-1} i.p.) and a polyethylene cannula was inserted into a femoral vein for administration of drugs. A second cannula was inserted into a carotid artery to measure systemic arterial blood pressure (BP).

Twenty minutes after the surgery, Paf (100 ng kg^{-1} min^{-1}) was infused intravenously for 10 min at a dose previously shown to induce extensive necrosis of the gastric mucosa (Wallace & Whittle, 1986a). Prior to the Paf infusion, the rats were pretreated with one of several inhibitors of arachidonic acid metabolism. The details of the route, dose and time of administration are listed in Table 1 and were selected on the basis of their previous use in this laboratory as inhibitors of arachidonic acid metabolism. Thirty minutes after the Paf infusion was terminated, the stomach was removed, opened along the greater curvature and immersed in neutral buffered formalin. After 10 min the stomach was pinned out, mucosal side up, to a wax block and immersed in fresh fixative. The stomach was photographed and gastric damage scored (0 to 3 scale) from these photographs in a blind manner (Rosam *et al.*, 1986). Samples of the fundus (corpus) were excised from standardized regions with a scalpel and were processed by routine techniques prior to embedding in paraffin. Sections (4 μm) were stained with haematoxylin and eosin, coded and examined under a light microscope.

Haematological analysis

In order to determine the time-course of Paf-induced haematological changes, experiments were performed in which blood samples (0.2 ml) were collected from the carotid cannula for determination of concentrations of white blood cells (WBC), erythrocytes (RBC), haemoglobin and platelets, as well as the haematocrit. As in the previous experiments, Paf (100 ng kg^{-1} min^{-1}) was infused for 10 min. Blood samples were taken 5 and 15 min prior to, 2.5 and 10 min after beginning, and 15 and 30 min after terminating the Paf infusion. The RBC and WBC counts and the determinations of haematocrit and haemoglobin concentration were performed using a Clay Adams Hematology Analyzer 5. The platelet counts were performed using a Clay Adams Ultra-Flo 100 whole blood platelet counter. Duplicate measurements were made for each blood sample and the coefficient of variation for these measurements was $< 2\%$.

The effects of pretreatment with prednisolone or BW755C on the Paf-induced changes in haematological parameters were also studied. Prednisolone (20 mg kg^{-1} s.c.) and BW755C (50 mg kg^{-1} p.o.) were administered 120 and 60 min prior to Paf, respectively, and blood samples were taken at the same times as in the experiments described above.

Plasma exudation

Rats were given 10 μCi of ^{125}I-human serum albumin (2.5 μCi mg^{-1}; Amersham) by intravenous injection 20 min before a 10 min infusion of Paf (100 ng kg^{-1} min^{-1}). Blood samples (0.3 ml) were collected from the carotid cannula into 1.5 ml Eppendorf tubes 5 and 15 min before, 2.5 and 10 min after the beginning and 15 and 30 min after terminating the Paf infusion. An aliquot portion (100 μl) of whole blood was transferred to a glass test tube and the remainder of the blood was centrifuged for 1 min (9000 *g*) in an Eppendorf benchtop centrifuge. The levels of [^{125}I]-albumin present in a 50 μl sample of the plasma and in the sample of whole blood were determined by counting the radioactivity in a γ-spectrometer.

Since some loss of albumin from the circulating blood occurs under control conditions, blood samples were collected from rats receiving an infusion of vehicle and the loss of radioactivity from these control experiments used to correct the values obtained in the Paf experiments.

The effects of pretreatment with prednisolone or BW755C on the Paf-induced plasma exudation were also studied. Prednisolone (20 mg kg^{-1} s.c.) and BW755C (50 mg kg^{-1} p.o.) were administered 120 and 60 min prior to Paf, respectively, and blood samples were taken at the same times as in the experiments described above.

Materials

Platelet-activating factor (1-O-alkyl-2-O-acetyl-*sn*-glyceryl-3-phosphorylcholine) was obtained from Sigma Chemical Company. An aliquot portion of the stock 2 mg ml^{-1} solution of Paf in chloroform was evaporated under a stream of nitrogen and was reconstituted in 0.25% (w/v) bovine serum albumin in 0.9% (w/v) saline when required. Paf was infused intravenously at a rate of 0.1 ml min^{-1}. Dexameth-

Table 1 Effects of inhibitors of arachidonic acid metabolism on the gastric mucosal damage induced by platelet-activating factor (Paf)

Pretreatment drug (n)	*Dose* ($mg\,kg^{-1}$) *and route*	*Time before Paf* (min)	*Gastric damage score* (0–3)
None (9)	—	—	3.0 ± 0
Dexamethasone (5)	2 i.v.	15	3.0 ± 0
Dexamethasone (4)	0.2 s.c.	120	1.6 ± 0.6*
Dexamethasone (12)	2 s.c.	120	1.2 ± 0.3**
Prednisolone (8)	20 s.c.	120	1.3 ± 0.4**
Indomethacin (8)	5 s.c.	45	2.5 ± 0.4
Acetylsalicylate (4)	10 i.v.	10	3.0 ± 0
Benzylimidazole (8)	50 s.c.	60	2.3 ± 0.3
BW755C (9)	50 p.o.	60	1.2 ± 0.4**

Gastric damage was scored in a randomized, blind manner on a 0 (normal) to 3 (severe hyperaemia and haemorrhage) scale. The damage score data is given as the mean ± s.e.mean of (n) experiments. Asterisks (*$P<0.05$; **$P<0.01$) denote groups which differ significantly from the group receiving Paf alone (Mann-Whitney U test).

asone sodium phosphate (Merck, Sharp & Dohme) was diluted in 0.9% saline to a concentration of $2\,mg\,ml^{-1}$ or $0.2\,mg\,ml^{-1}$ before use. Prednisolone 21-sodium succinate (Sigma), 1-benzylimidazole fumarate (Wellcome Research Laboratories), acetylsalicyclic acid (Monsanto) and BW755C (W.R.L.; 3-amino-[m-(trifluoromethyl)-phenyl]-2-pyrazoline) as the hydrochloride were dissolved in 0.9% saline immediately before use. Indomethacin (Sigma) was dissolved in 1.25% sodium bicarbonate immediately before use.

Statistical analysis

Results are expressed as mean ± s.e.mean. Comparisons between groups of parametric data were made by Student's t test for unpaired data. Comparisons between groups of non-parametric data were made by the Mann-Whitney U-test. With all analyses, an associated probability (P value) of 5% or less was considered as significant. A linear regression analysis was performed on the data from the Paf, BW755C and prednisolone groups to determine if there was a correlation between gastric damage score data and the final erythrocyte concentrations.

Results

Gastric mucosal damage

Intravenous infusion of Paf for 10 min induced extensive gastric hyperaemia and haemorrhage, with all rats in this group ($n = 9$) being assigned the maximum gastric damage score of 3 (see Table 1). This hyperaemia was located almost exclusively in the fundic region of the stomach along the crests of rugal folds. The border of the fundus and antrum, along the lesser curvature, was also a prominent site of mucosal hyperaemia. Histologically, the gastric damage was characterized by extensive vascular congestion, with focal regions of mucosal necrosis and haemorrhage. Accumulations of neutrophils and neutrophil aggregates were evident in the arterioles and venules at the base of the mucosa and in the submucosa in all of the treatment groups. While no quantification was performed, the various drug pretreatments did not have any noticeable effect on the numbers of such neutrophil aggregates. Other histological features of the Paf-induced damage were identical to those described previously (Rosam *et al.*, 1986; Wallace & Whittle, 1986a).

Of the various drug pretreatments, only the corticosteroids, when given 2 h before Paf, and BW755C significantly reduced the gastric damage score (Table 1). In these groups, gastric damage was limited to diffuse hyperaemia or, in some cases, a few patches of hyperaemia and haemorrhage, while in some animals the gastric mucosa appeared normal. The histological assessment of damage confirmed the mascroscopic observations. Thus, where hyperaemia was observed macroscopically, mucosal vasoconges-tion was apparent histologically. The few stomachs that were given a score of 0 on the macroscopic assessment were confirmed histologically to be devoid of damage.

Pretreatment with indomethacin, acetyl salicylic acid, benzylimidazole and the acute intravenous administration of dexamethasone ($2\,mg\,kg^{-1}$) had no significant effect on the gastric damage score, or on the histological appearance of the gastric mucosa.

Arterial blood pressure

The infusion of Paf caused a rapid, biphasic fall in BP (Figure 1). The initial fall in BP of 78 ± 2 mmHg

began immediately after starting the Paf infusion and was complete within 10 s. Invariably, there was a further fall in BP of 10 ± 2 mmHg 4–5 min after the Paf infusion was started. During the 30 min after completion of the Paf infusion, the BP very gradually increased, but by the end of the experiment had only recovered to 39 ± 7% of basal levels.

Of the various pretreatments, only BW755C, acetylsalicyclic acid and benzylimidazole had a significant effect on the resting BP (Figure 1), each causing a slight hypotension. None of the drugs signifcantly affected the fall in BP induced by Paf, although the recovery of BP following the Paf infusion was significantly accelerated by several of the drugs. In the groups in which the corticosteroids were given 2 h before Paf, the BP rapidly recovered to basal levels. In these groups, the BP values were significantly ($P < 0.001$) higher than the corresponding control BP at each point after the infusion of Paf. In the group given acute intravenous dexamethasone as well as in the indomethacin and BW755C groups, the recovery of BP was somewhat more rapid, but did not fully recover to basal levels and was not significantly different from the Paf control group at the end of the experiment. Pretreatment with acetylsalicylic acid or benzylimidazole had no effect on the Paf-induced changes in BP.

Haematological analysis

In basal blood samples, the mean haematrocrit was 42.4 ± 1.5%, and the mean RBC, WBC, haemoglobin and platelet concentrations were, respectively, $6.4 \pm 0.2 \times 10^6\,mm^{-3}$, $5.7 \pm 0.2 \times 10^3\,mm^{-3}$, $13.2 \pm 0.3\,g\,dl^{-1}$ and $700 \pm 24 \times 10^3\,mm^{-3}$. These levels were not significantly affected by pretreatment of the rats with prednisolone (20 mg kg^{-1} s.c.) or BW755C (50 mg kg^{-1} p.o.). The infusion of Paf caused a transient leukopenia ($P < 0.001$) followed by a profound haemoconcentration (Figure 2). In samples taken at the end of the Paf infusion and thereafter, the RBC, haemoglobin and platelet concentrations and the haematocrit were increased by 25–45% above ($P < 0.001$) those in samples taken before Paf.

Pretreatment with either BW755C or prednisolone did not significantly alter the Paf-induced leukopenia (Table 2), but did cause a significant reduction ($P < 0.05$) in the Paf-induced increases in RBC, haemoglobin and platelet concentrations, as well as

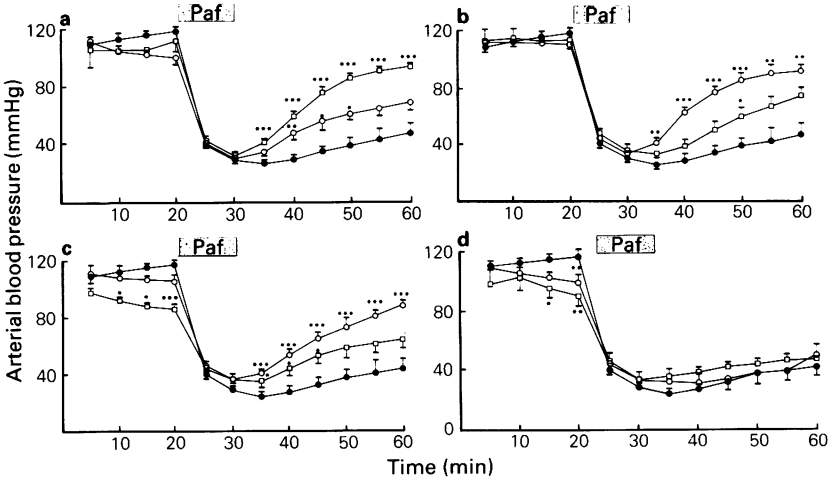

Figure 1 The effects of platelet-activating factor (Paf) and the various drug pretreatments on systemic arterial blood pressure. The data for the Paf control group is shown in each panel (●). Paf (100 ng kg^{-1} min^{-1}) was infused intravenously during the 10 min period indicated by the stippled bar. The results are shown as the mean with vertical lines indicating s.e.mean, except on some points where, for the sake of clarity, the s.e.mean has been omitted. The sizes of the samples for each group are listed in Table 1. Asterisks denote points at which the pretreated group differs significantly (*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$) from the Paf control group. (a) Rats were pretreated with (□) dexamethasone (2 mg kg^{-1} s.c.) 2 h before Paf or (○) indomethacin (5 mg kg^{-1} s.c.) 1 h before Paf. (b) Rats were pretreated with (○) dexamethasone (0.2 mg kg^{-1} s.c.) 2 h before Paf or (□) dexamethasone (2 mg kg^{-1} i.v.) 15 min before Paf. (c) Rats were pretreated with (○) prednisolone (20 mg kg^{-1} s.c.) 2 h before Paf or (□) BW755C (50 mg kg^{-1} p.o.) 1 h before Paf. (d) Rats were pretreated with (○) acetylsalicylic acid (10 mg kg^{-1} i.v.) 10 min before Paf or (□) benzylimidazole (50 mg kg^{-1} s.c.) 1 h before Paf.

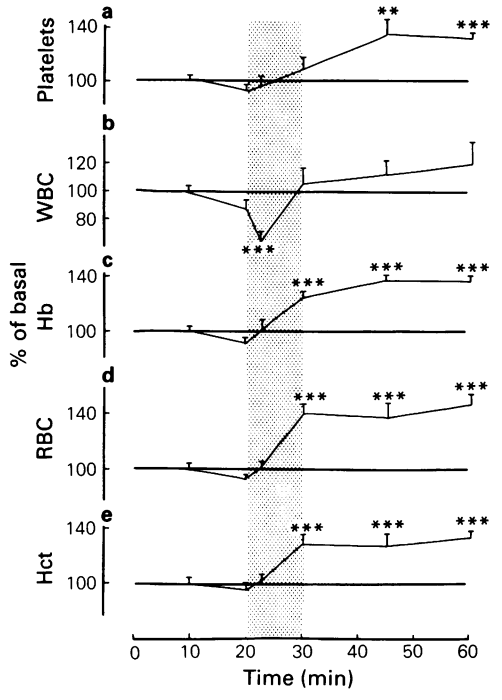

Figure 2 Effects of platelet-activating factor (Paf) on (e) haematrocit (Hct) and the concentrations of (d) erythrocytes (RBC), (b) leukocytes (WBC), (c) haemoglobin (Hb) and (a) platelets in the blood. Paf was administered intravenously (100 ng kg^{-1} min^{-1}) during the 10 min period indicated by the stippled area. Results are expressed as the mean % of basal levels ($n = 6$; see text for actual basal values). Vertical lines show s.e.mean. Asterisks denote points which are significantly different (**$P < 0.01$; ***$P < 0.001$) from the basal levels.

haematocrit. In these groups and the Paf control group there was a highly significantly correlation ($r = 0.78$; $P < 0.01$) between the gastric damage score and the extent of haemoconcentration (expressed as the erythrocyte concentration as % of basal).

Plasma exudation

There was no detectable change in the blood levels of radiolabelled albumin in the samples taken 2.5 min after the beginning of the infusion of Paf (Figure 3). However, in the samples taken at the end of the Paf infusion, there was a significant decrease (27 ± 3%; $P < 0.05$) in the concentration of radioactive material present. There was a further reduction in the concentration of radiolabelled albumin in the samples of blood taken during the 30 min following the Paf infusion. Since there was only a small loss (< 10%) of [^{125}I]-albumin from the plasma during the experiments, the decrease in circulating levels of

Table 2 Effects of BW755C and prednisolone on the leukopenia and haemoconcentration induced by platelet-activating factor (Paf)

Group (n)	*Leukopenia* (%)	*Haemoconcentration* (%)
Control (6)	39.6 ± 6.3	48.4 ± 6.5
Prednisolone (6)	36.9 ± 5.0	12.9 ± 2.8***
BW755C (6)	26.9 ± 4.2	13.1 ± 1.7***

Paf (100 ng kg^{-} min^{-1}) was infused intravenously for 10 min. Leukopenia was determined as the concentration of white blood cells in the blood sample taken 2.5 min after beginning the Paf infusion as a percentage of the basal concentration. Haemoconcentration was determined as the concentration of erythrocytes in the final blood sample of the experiment as a percentage of the basal concentration. Prednisolone (20 mg kg^{-1} s.c.) was administered 120 min before Paf while BW755C (50 mg kg^{-1} p.o.) was administered 60 min before Paf. Asterisks denote groups which differ significantly (***$P < 0.001$) from the control group (Student's t test).

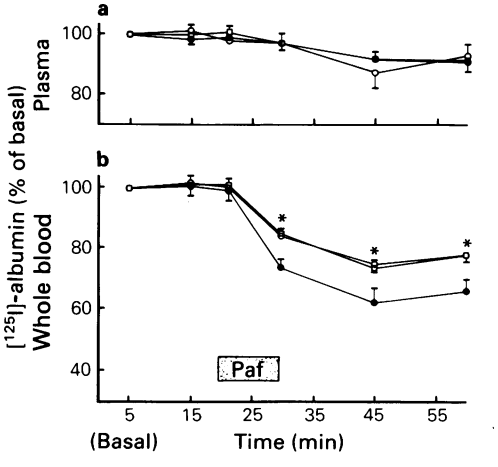

Figure 3 The effects of pretreatment with (○) BW755C (50 mg kg^{-1} p.o.) or (□) prednisolone (20 mg kg^{-1} s.c.) on platelet-activating factor (Paf)-induced loss of ^{125}I-human serum albumin from plasma (a) and whole blood (b). Paf was administered (100 ng kg^{-1} min^{-1}) during the 10 min period indicated by the stippled bar. Results are expressed as the mean % of basal levels ($n = 4$ per group). Vertical lines show s.e.mean. Asterisks denote points where both th BW755C and prednisolone groups differ significantly (*$P < 0.05$) from the Paf control group (●).

radiolabelled albumin can be attributed to plasma exudation. Pretreatment with prednisolone or BW755C caused a significant reduction ($P < 0.05$) in the Paf-induced plasma exudation (Figure 3).

Discussion

These results confirm our previous observations that platelet-activating factor induces extensive gastric mucosal necrosis (Rosam *et al.*, 1986; Wallace & Whittle, 1986a). The present study further indicates that such damage can be significantly reduced by pretreatment with dexamethasone, prednisolone or BW755C. A role for cyclo-oxygenase products of arachidonic acid in the mechanism of Paf-induced ulceration seems unlikely, since pretreatment with either indomethacin or acetylsalicylic acid had no significant protective effect. The lack of a role for the cyclo-oxygenase product, thromboxane A_2, is further supported by the failure of a thromboxane synthase inhibitor, 1-benzylimidazole (Whittle *et al.*, 1981), to provide any protection against Paf-induced gastric damage. Hsueh *et al.* (1986) recently found that infusion of an extremely high dose of prostaglandin E_1 (PGE_1, 2 mg kg^{-1} min^{-1} for 2 h) ameliorated the small bowel necrosis induced by high doses of Paf. They also reported an augmentation of Paf-induced small bowel necrosis by indomethacin which, along with the observations on PGE_1, led them to conclude that vasodilator prostaglandins play a defensive role against Paf-induced damage. It is not yet known whether lower, more appropriate doses of exogenous prostaglandins will exert a protective effect against Paf-induced gastric damage.

While a role for cyclo-oxygenase products of arachidonic acid in Paf-induced gastric damage is unlikely, a role for lipoxygenase products cannot be excluded. The drugs which significantly reduced the gastric damage score can all inhibit the synthesis of 5-lipoxygenase products. BW755C has been shown to inhibit 5-lipoxygenase activity *in vivo* in inflammatory exudates (Salmon *et al.*, 1983) while corticosteroids inhibit the release of arachidonic acid through inhibition of phospholipase A_2 (Flower & Blackwell, 1979; Blackwell *et al.*, 1980). Interestingly, dexamethasone did not reduce gastric damage when given only 15 min before Paf but did if it was given 2 h before Paf. This observation is consistent with the effects of dexamethasone being mediated through the time-dependent biosynthesis of a lipocortin (Blackwell *et al.*, 1980).

Beneficial effects of corticosteroids and lipoxygenase inhibitors against damaging effects of Paf have been observed in other models. For example, Myers *et al.* (1983) reported that cortisone acetate exerted a highly protective effect against Paf-induced toxicity in mice. Using a similar animal model, Young *et al.* (1985) showed that dexamethasone (given at least 1.5 h before Paf) and several putative lipoxygenase inhibitors (phenidone, nordihydroguaiaretic acid and diphenyldisulphide), as well as BW755C, provided nearly complete protection against the mortality associated with Paf-induced shock. Hsueh *et al.* (1986) recently found that the leukotriene D_4 antagonist, FPL 55712, as well as the non-specific lipoxygenase inhibitor, nordihydroguaiaretic acid, reduced the extent of Paf-induced damage to the rat small bowel.

A role for lipoxygenase products as mediators of gastric mucosal damage has not yet been established, but there is some evidence for such an action. For instance, a potent vasoconstrictor action of leukotriene C_4 (LTC_4) in the gastric submucosal microvessels, which could predispose the mucosa to necrosis, was recently demonstrated (Whittle *et al.*, 1985). BW755C was shown to reduce ethanol-induced gastric mucosal damage, which may reflect its inhibitory action on lipoxygenase enzymes (Wallace & Whittle, 1985). Furthermore, Peskar *et al.* (1986) showed that the rat gastric mucosa released LTC_4 in response to oral administration of ethanol and that the ethanol-induced gastric damage was inhibited by nordihydroguaiaretic acid.

It is conceivable that Paf-induced neutrophil aggregation and degranulation leads to the release of leukotrienes (Chilton *et al.*, 1982), which then contribute to the congestion of the microvessels in the gastric mucosa through vasoconstriction. Consistent with this hypothesis is our observation that Paf-induced leukopenia preceded the haemoconcentration. Furthermore, Smith & Bowman (1982) observed that lipoxygenase inhibitors prevented Paf-induced neutrophil degranulation *in vitro*, while cyclo-oxygenase inhibitors had no effect. However, the potential role of local vasoconstriction, either by Paf or by a secondary mediator, is doubtful since a recent study using *in vivo* microscopy techniques showed that Paf had no effect on the diameter of submucosal venules or arterioles or of mucosal capillaries (Whittle *et al.*, 1986). Neutrophil aggregates could contribute to the mucosal vasocongestion induced by Paf by occluding microvessels (Wallace & Whittle, 1986a). Neutropenia following Paf infusion to rats has previously been reported (McManus *et al.*, 1980) and it was suggested that this was a result of aggregation of neutrophils and their subsequent sequestration in the microvasculature (Camussi *et al.*, 1983). However, since some accumulation of neutrophils was still evident in samples of protected mucosae, histological data on neutrophil aggregates will be required to address this more fully.

As has been demonstrated previously (Bessin *et al.*, 1983; Doebber *et al.*, 1984), infusion of Paf resulted in of both BW755C and prednisolone to significantly reduce Paf-induced plasma exudation and haemocon-

centration is supportive of a role for these processes in the aetiology of Paf-induced gastric damage, as is the highly significant correlation between gastric damage score and degree of haemoconcentration. Our results clearly show that the prolonged hypotension alone did not account for the gastric damage. Pretreatment with BW755C, indomethacin and acute intravenous administration of dexamethasone had similar effects on the recovery of BP after Paf, but of these treatments only BW755C significantly reduced gastric damage.

The mechanism by which corticosteroids and BW755C reduce plasma exudation and haemoconcentration is not clear, although their inhibitory actions on lipoxygenase and leukotriene synthesis may again be relevant. Leukotrienes C_4 and D_4 have been shown to increase vascular permeability (Dahlen *et al.*, 1981), so it is possible that they could mediate Paf-induced effects on vascular permeability. A further possibility is that Paf infusion leads to endogenous Paf release, as has been shown previously (Bourgain *et al.*, 1985), and that the beneficial effects of BW755C or the corticosteroids can be attributed to an inhibition of this release. Paf biosynthesis is dependent on the significant haemoconcentration, probably as a consequence of increased vascular permeability. Coupled with the prolonged hypotension, this haemoconcentration could account for the sluggishness of mucosal blood flow caused by Paf (Whittle *et al.*, 1986) and thereby predispose the mucosa to damage. The ability activity of phospholipase A_2 (Mencia-Huerta & Benveniste, 1979) and the release of the precursor lyso-Paf can be inhibited both *in vivo* and *in vitro* by corticosteroids (Parente & Flower, 1985; Parente *et al.*, 1986).

We have previously proposed that Paf is an endogenous mediator of gastrointestinal ulceration in septic shock (Rosam *et al.*, 1986). If our hypothesis is correct, then drugs which interfere with the ulcerogenic actions of Paf may be beneficial in the treatment of septic shock. Indeed, corticosteroids are widely used clinically in the treatment of sepsis and have also been shown to prevent the mortality associated with endotoxin-induced shock in several animal models (Nicholson, 1982). Furthermore, we have recently demonstrated that CV-3988, a Paf-antagonist, inhibited the gastrointestinal ulceration induced by endotoxin or Paf (Wallace & Whittle, 1986b). However, further studies are required to determine the possible role of lipoxygenase products in the gastrointestinal ulceration associated with septic shock or with the intravenous infusion of Paf.

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