Effects of PAF and BN 52021 on cardiac function and regional blood flow in conscious rats

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SIRÉN, ANNA-LEENA, AND GIORA FEUERSTEIN. *Effects of PAF and BN 52021 on cardiac function and regional blood flow in conscious rats.* Am. J. Physiol. 257 (Heart Circ. Physiol. 26): H25–H32, 1989.—The effect of intravenous injections (0.1–3 nmol/kg) of platelet-activating factor (PAF) on blood pressure, heart rate, cardiac output, and blood flow (hindquarter, renal, mesenteric) were studied in conscious rats. PAF decreased blood pressure and total peripheral reistance (TPR) but increased heart rate; cardiac output was reduced by the highest dose. Low doses of PAF increased blood flow and decreased vascular resistance in all vascular beds, whereas high doses reduced mesenteric blood flow in part by increasing mesenteric vascular resistance. The hypotensive and cardiac effects of PAF were blocked by intravenous infusions of the selective PAF-receptor antagonists, 15 mg/kg BN 52021 and 1 mg/kg SDZ 63-441. BN 52021 also attenuated the hindquarter and renal responses to PAF, but the mesenteric responses remained relatively unchanged. The results indicate that PAF is a potent vasodilator of mesenteric > hindquarter = renal vessels at low doses and a cardiac depressant at high doses. A therapeutic role for the PAF antagonists BN 52021 and SDZ 63-441 is suggested in endotoxemia, anaphylaxis, and other disease states in which increased release of PAF contributes to key hemodynamic derangements.

blood pressure; heart rate; cardiac output; vascular resistance; mesenteric blood flow; renal blood flow; hindquarter blood flow; platelet-activating factor-induced shock; SDZ 63-441; alprazolam

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PLATELET-ACTIVATING FACTOR (PAF) is a glycerophospholipid produced and released from stimulated cells such as basophils, macrophages, platelets, polymorphonuclear neutrophils, endothelial cells, and mast cells (29). Systemic administration of PAF induces hypotension in various animal species (for review see Ref. 8). Decreases in arterial pressure, cardiac output, and total peripheral resistance after PAF administration have been reported in the anesthetized rat and dog (12, 22, 30). Although the effects of PAF on gross cardiovascular variables have been repeatedly reported, only a few reports to date have studied the discrete organ blood flow changes after PAF administration in intact animals. In the anesthetized dog, PAF decreased renal blood flow (2, 30), whereas vasodilation was reported in the femoral artery (27). In the anesthetized domestic pig, PAF produced a dual effect on coronary circulation: vasodilation followed by vasoconstriction (9). Decreases in blood flow to brain,

heart, kidneys, lungs, and spleen in response to PAF infusions were described in the anesthetized spontaneously hypertensive rat (SHR, Ref. 12). However, none of the previous studies attempted to examine the relative contribution of cardiac output and organ blood flow in PAF-induced circulatory shock. Furthermore, in the species studied previously, i.e., dog, pig, guinea pig, and rabbit, PAF is a potent aggregator and activator of platelets and white blood cells; vasoactive substances (e.g., thromboxane A_2 and leukotrienes) released from these cellular elements were shown to mediate most of the PAF effects (8, 9, 13, 18, 27). Rat platelets like those of humans, are relatively resistant to PAF actions (8, 23), and therefore the rat could serve as a useful model to study blood vessel responses to PAF independent of platelet aggregation.

The present study examined the systemic and regional hemodynamic changes that accompany the hypotensive effect of PAF in conscious rats. In addition, the influences of specific PAF-receptor antagonists, BN 52021 (4), SDZ 63-441 (13), and the benzodiazepine alprazolam (16), on various blood vessel and cardiac responses to PAF were also studied.

MATERIALS AND METHODS

Male Sprague-Dawley rats (300–310 g) were purchased from Taconic Farms (Germantown, NY) and kept at 22°C and in a 12-h light-dark cycle. After surgical operations, the rats were housed individually in plastic cages (21 × 27 × 16 cm, W × L × H) with food and water ad libitum and monitored daily for adequate recovery from surgery.

Measurement of cardiac output. The rats were anesthetized with an intramuscular injection of 130 mg/kg ketamine and 1.3 mg/kg acepromazine, and PE-50 catheters were inserted into femoral vessels. The catheters were tunneled under the skin and exited at the nape of the neck. An incision was made at the midline of the neck from the cricoid to the clavicle, and a PE-50 catheter was inserted into the right atrium through the external jugular vein. The left common carotid artery was exposed and ligated, and a thermistor (MX2-780-33 model THMP f 1.5, Teflon reusable, Columbus Instruments, OH) was advanced through the carotid into the ascending aorta (placement above the aortic valve was confirmed in each animal at the end of the experiment and by the shape of the dilution curve before the probe was finally

sutured to the neck muscles). The jugular vein catheter and the thermistor were tunneled under the skin to the nape of the neck. All catheters and the probe wire were secured by a soft spring wire attached to the animal's neck with an adhesive collar. Twenty-four hours after the surgery, the arterial line was connected to a pressure transducer (Narco Bio-Systems model RP 1500i) coupled to a strain gauge coupler (Narco Bio-Systems type 7032). Blood pressure (mean, systolic, diatolic, pulse) and heart rate were continuously recorded on a Narcotrace 80 computerized physiograph and sampled automatically at 30- to 60-s intervals by a Northstar-Hazeltine computer.

The cardiac output was measured by thermodilution technique as previously described (25). In brief, the thermistor was attached to the computerized Cardiomax II (CMX2-780-k with microprobe option R, Columbus Instruments). The dead space of the venous line was first flushed with 0.05 ml of 0.9% (wt/vol) NaCl (saline) at room temperature (22°C); after a brief stabilization period an additional injection of 0.2 ml normal saline (22°C) was rapidly injected using a 1-ml syringe. For cardiac output measurement a control period of 15 min included two or three cardiac output recordings to test for consistency and placement of the probe. During this period control values for blood pressure and heart rate were also collected. The timer of the automatic data collection was started, and data points were collected immediately before and 30 s, 2, 5, 10, 20, and 30 min after the PAF injection. Total peripheral resistance index (TPRI) was calculated by dividing the mean arterial pressure by the cardiac output; values of cardiac output and TPRI were further indexed per unit of weight (kg).

Measurement of organ blood flow. The directional pulsed-Doppler method was selected to measure organ blood flow in the hindquarter, renal, and mesenteric vessels. Although it does not allow quantitative blood flow monitoring, this method is superior compared with other available techniques; because it can be used chronically in conscious animals, it allows continuous on-line recording of blood flow and detections of instantaneous and transient changes in blood flow within seconds after the drug administration. Haywood and co-workers (14) have shown that the velocity signals recorded from the Doppler flow probes are directly and reliably proportional to changes in true volume flow measured by electromagnetic flowmetry.

The rats were anesthetized with ketamine-acepromazine as described in *Measurement of cardiac output.* A midline laparotomy was made, and the left renal and superior mesenteric arteries and lower abdominal aorta above its bifurcation were carefully isolated under a dissecting microscope. Doppler flow probes (Valpey-Fisher, Hopkinton, MA) were then loosely sutured around each vessel. The insulated wire leads were fixed to the back muscles, tunneled under the back skin to exit at the neck, and soldered to a receptacle that was then attached to the skull with small screws and dental acrylic. The animals were allowed to recover from the surgery for 7 days. Twenty-four hours before the experiment, the rats were reanesthetized with halothane (2% in O_2), and the femoral artery and vein were catheterized with PE-

50 tubing. The catheters were tunneled under the back skin, exited at the nape of the neck, and secured by a soft spring wire as described above.

On the day of the experiment, the arterial catheter was connected to a pressure transducer (Narco), and blood pressure and heart rate were continuously recorded on the Narcotrace 80 physiograph. A cable connecting the blood flow recepticle and the Doppler flowmeter (University of Iowa, Bioengineering Facility, model 545C-4) was attached to the animal, and the mean blood flow was continuously recorded on the physiograph. Vascular resistance was calculated by dividing the mean arterial pressure by blood velocity (Doppler shift in kHz) as earlier described (14, 25). Changes in blood flow and vascular resistance are expressed as a percent of control values.

Drugs used. Pure synthetic 1-O-hexadecyl-2-acetyl-sn-glycero-3-phosphorylcholine (PAF) was kindly provided by Dr. F. Snyder, Oak Ridge Associated Universities, Oak Ridge, TN. PAF was dissolved in saline to a stock solution of 100 nmol/ml, which was kept frozen (−70°C) until used, and diluted to a final volume of 100 μl per injection. Pure powdered BN 52021 [9H1,7a-(epoxymethano)-1H,6aH-cyclopenta[c]furo[2,3-b]furo-[3′,2′,3,4] cyclopenta[1,2-d]furan-5,9,12-(4H)trione, 3-tert-butylhexahydro-4,-7b,-11hydroxy-8methyl] was kindly provided by Dr. P. Braquet, IHB Research Laboratories, France. For administration to the rat, BN 52021 was first dissolved in 50 μl of 50% dimethyl sulfoxide (DMSO) and further diluted with 1 ml of normal saline. The pH was adjusted to the range of 7–8 by titration with 10 N NaOH. Alprazolam (kindly provided by Upjohn, Kalamazoo, MI) was first dissolved in absolute ethanol and propyleneglycol and further diluted in saline. The concentrations of the above mentioned chemicals in the final solution were 10 ethanol, 40 propylene-glycol, and 50% saline. *Cis*-(±)-1-[2-[hydroxy[[tetra-hydro-5-[(octadecylaminocarbonyl)oxy]methyl]-furan-2-yl]methoxyphosphinyloxy]ethyl]quinolinium hydroxide (SDZ 63-441, Sandoz Research Institute, East Hanover, NJ) was dissolved in saline immediately before injection.

Increasing doses of 0.1–3 nmol/kg PAF were injected intravenously at 30- to 90-min intervals. The effect of the preceding dose had completely subsided before the administration of each new dose; 15 mg/kg BN 52021 was injected 15 min before 0.3–3 nmol/kg PAF, 3 mg/kg alprazolam was injected 20 min before 1–3 nmol/kg PAF, and SDZ 63-441 was injected 2 min before 1 nmol/kg PAF.

Statistical analysis of data. Data in text and figures are represented as means ± SE for the given number of rats. One-way analysis of variance (ANOVA) followed by Student-Newman-Keuls test or Kruskal-Wallis test (28) was used for statistical analysis of the data. A significant difference was accepted at $P < 0.05$.

RESULTS

Effects of PAF on systemic hemodynamic variables. The base-line blood pressure and heart rate were not influenced by repeated injections of PAF (Fig. 1). The PAF
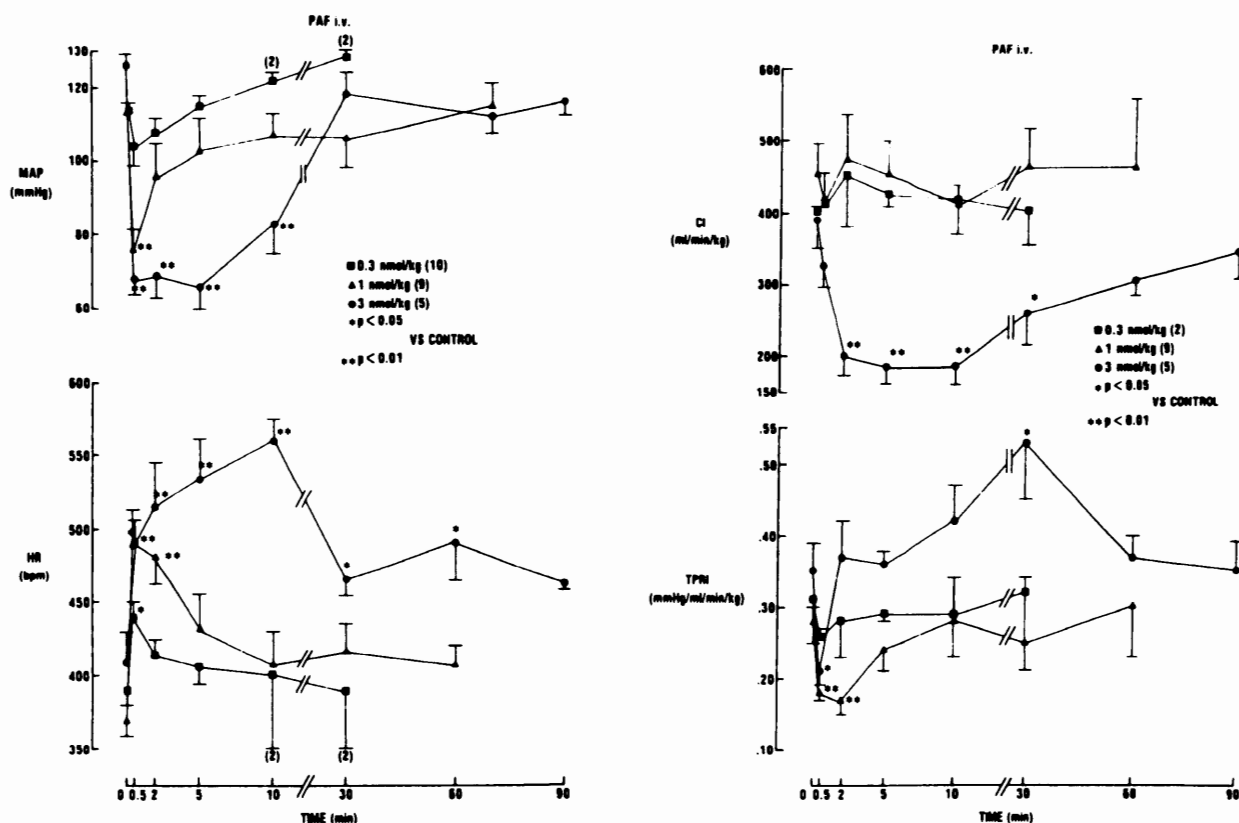
FIG. 1. Dose-response effect of PAF on mean arterial pressure (MAP), heart rate (HR), cardiac index (CI), and total peripheral resistance index (TPRI) in conscious rats. Increasing doses of 0.3–3 nmol/kg PAF were injected intravenously at 60- to 90-min intervals. Values indicate means ± SE. No. of rats is given in parentheses. Asterisks denote statistical significance from control by Kruskal-Wallis test.

doses up to 1 nmol/kg had no effect on hematocrit. The hematocrit before the 1-nmol/kg dose was 43 ± 2%, and 60 min after this dose it was 44 ± 2%. PAF (1- to 3-nmol/kg) induced a dose-related decrease in mean arterial pressure (MAP) and increased heart rate (Fig. 1). The maximum hypotensive and tachycardic effects after each dose of PAF were achieved in 30–60 s and subsided in 5–15 min. The lower doses (0.1–0.3 nmol/kg) had no effect on blood pressure (maximum changes of −2 ± 3 and −10 ± 4 mmHg, respectively), but the 0.3-nmol/kg dose induced a significant increase in heart rate (Fig. 1).

The base-line values of cardiac index before PAF administration ranged from 380 to 458 ml·min⁻¹·kg⁻¹. The range of the cardiac index values in the present study is well in agreement with our previous findings (25) as well as those of others utilizing the thermodilution method (21). The 1-nmol/kg dose of PAF decreased TRPI with no consistent effect on cardiac index; in five of 12 animals, PAF increased cardiac index (+91 ± 21 ml·kg⁻¹·min⁻¹), whereas the other seven animals reacted to the PAF injections with a decrease in cardiac index (−104 ± 22 ml·min⁻¹·kg⁻¹). The highest dose produced a marked decrease in cardiac index (Fig. 1). The fall in TPRI became apparent with the maximum hypotensive response 30 s after PAF administration and subsided in 5 min. The 3-nmol/kg dose of PAF first transiently decreased and then increased TPRI.

Effect of PAF antagonists on systemic hemodynamic

responses to PAF. Fifteen milligrams per kilogram of BN 52021 or 1 mg/kg SDZ 63-441 had no effect on the basal values of blood pressure, heart rate, cardiac index, or TPRI, whereas alprazolam significantly decreased MAP and increased heart rate resulting in a significantly lower MAP and a higher heart rate before PAF administration in the alprazolam-treated group (Table 1).

BN 52021 totally blocked the hypotensive effect of the 1-nmol/kg dose of PAF (Table 2) and effectively attenuated but never completely abolished the decrease in MAP produced by the 3-nmol/kg dose (Fig. 2). The increase in heart rate induced by PAF was blocked by

TABLE 1. *Base-line values of gross cardiovascular variables before PAF administration in BN 52021-, SDZ 63-441-, or alprazolam-treated and control rats*

Group	n	MAP, mmHg	HR, beats/min	CI, ml·min⁻¹·kg⁻¹	TPRI, mmHg·ml⁻¹·min·kg
Control	8–14	118±4	376±12	370±47	0.30±0.03
BN 52021	9	112±3	414±20	451±28	0.26±0.02
SDZ 63-441	4	118±6	411±43	349±17	0.34±0.01
Alprazolam	10	103±3*	412±14*	NT	NT

Values are means ± SE; n, no. of rats. MAP, mean arterial pressure; HR, heart rate; CI, cardiac index; NT, not tested; TPRI, total peripheral resistance index. Measurements were made 20 min after iv injection of saline, 15 mg/kg BN 52021, or 3 mg/kg alprazolam, and 2 min after 1 mg/kg SDZ 63-441. This time point denotes situation immediately before administration of 1st dose of PAF. * Significance from control group, P < 0.05 (Dunnett's test).

TABLE 2. *Modulation of the hemodynamic responses to 1 nmol/kg PAF by the PAF antagonists BN 52021, SDZ 63-441, and alprazolam*

Group	n	MAP, mmHg	HR, beats/min	CI, $ml \cdot min^{-1} \cdot kg^{-1}$	TPRI, $mmHg \cdot ml^{-1} \cdot min \cdot kg$
PAF	12	−35±5	+89±19	+14±21	−0.11±0.03
PAF after BN 52021	4	+8±5*	+29±14	+104±30†	−0.02±0.01
PAF after SDZ 63-441	4	0±3*	+24±23	+66±11	−0.05±0.01
PAF after alprazolam	10	−46±5	+104±24	NT	NT

Values are means ± SE and represent maximum changes 30 s to 2 min after PAF administration. MAP, mean arterial pressure; HR, heart rate; CI, cardiac index; TPRI, total peripheral resistance index; NT, not tested. Intravenous injections of 15 mg/kg BN 52021 or 3 mg/kg alprazolam were given 20 min before PAF. SDZ 63-441 was administered 2 min before PAF. Compared with PAF alone: * $P < 0.01$; † $P < 0.05$ (Dunnett's test).

BN 52021 (Fig. 2, Table 2). BN 52021 totally abolished the cardiac depressant effect of the 3-nmol/kg dose of PAF but failed to attenuate the PAF-induced decrease in TPRI (Fig. 2). After BN 52021, the 1-nmol/kg dose of PAF increased cardiac index in all animals. In the BN 52021-treated rats, the peak increase in cardiac index (+104 ± 30 ml·kg⁻¹·min⁻¹, $n = 4$, $P < 0.01$ vs. control) was reached 2 min after PAF injection, and the cardiac index was still significantly elevated (+67 ± 45 ml· min⁻¹·kg⁻¹, $n = 4$, $P < 0.05$ vs. control) 10 min after the PAF injection.

A 1-mg/kg intravenous injection of SDZ 63-441 blocked the hypotensive effect of 1 nmol/kg PAF and attenuated the tachycardia (Table 2). After SDZ 63-441 treatment, PAF increased cardiac index in all animals. The peak increase in cardiac index by PAF in the SDZ 63-441-treated rats was +66 ± 11 ml·min⁻¹·kg⁻¹ ($n = 4$, $P < 0.01$ vs. control).

A 3-mg/kg intravenous injection of alprazolam had no effect on the hypotensive and tachycardiac responses to 0.3–3 nmol/kg PAF (Table 2).

Effects of PAF on organ blood flow and vascular resistance. Figure 3 demonstrates an original tracing of the effect of PAF on arterial pressure and regional blood flow. The 0.3-nmol/kg dose of PAF, which had no significant effect on MAP, induced a profound increase in mesenteric blood flow with no effect on renal or hindquarter vessels (Figs. 3 and 4). The higher dose (1 nmol/kg) decreased arterial pressure and renal blood flow, increased hindquarter blood flow, and initially increased but then decreased mesenteric blood flow. The maximum responses were reached 30–60 s after the injection and subsided within 5 min after the low doses, whereas after the higher doses, blood flow in all three vascular beds was decreased over a period of 5–10 min.

PAF (0.1–1 nmol/kg) decreased mesenteric vascular resistance in a dose-related manner (Fig. 4), whereas the highest dose (3 nmol/kg) induced primarily mesenteric vasoconstriction. The renal vascular resistance was significantly decreased after the 1- and 3-nmol/kg doses of PAF, with the maximum decrease (−38 ± 6%, $P < 0.01$
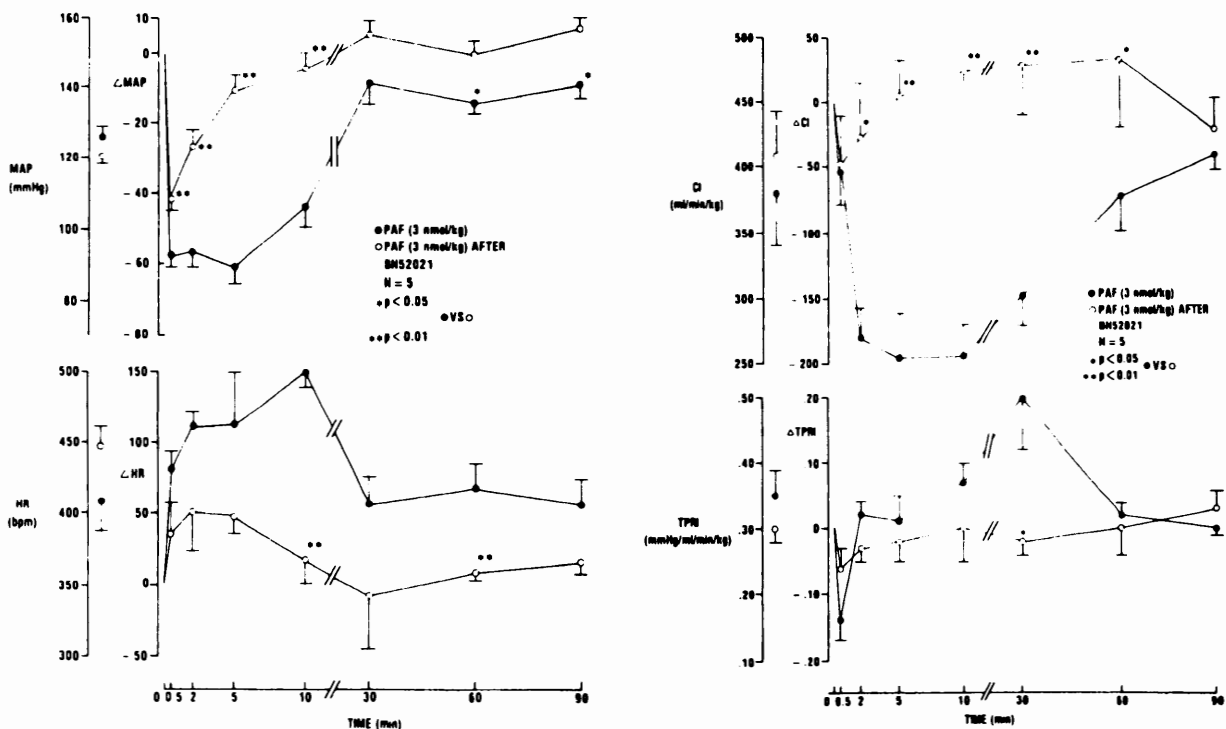

FIG. 2. Effect of 3 nmol/kg PAF on mean arterial pressure (MAP), heart rate (HR), cardiac index (CI), and total peripheral resistance index (TPRI) in control rats and rats intravenously injected with 15 mg/kg BN 52021 20 min before PAF administration. *Left*: each graph represents base-line values of MAP, HR, CI, or TPRI in control and BN 52021-treated rats. *Right*: each graph demonstrates changes of MAP and HR calculated from these base-line values immediately before PAF administration. Values indicate means ± SE. Asterisks indicate statistical significance between groups by Kruskal-Wallis test.
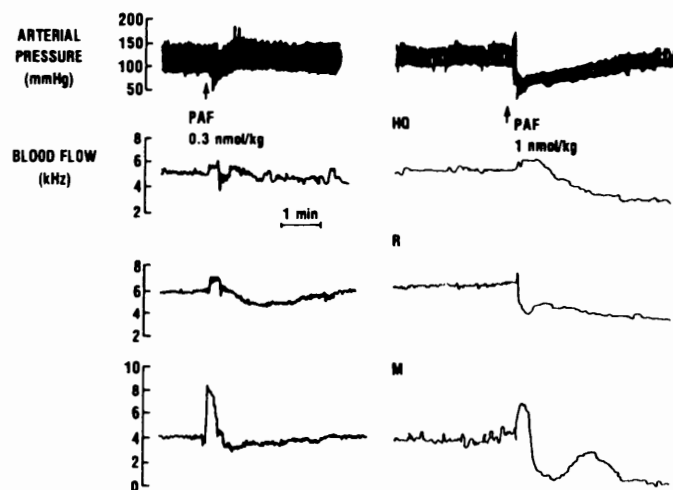
FIG. 3. A representative chart recording of effect of 0.3–1 nmol/kg PAF on arterial pressure (AP) and organ blood flow in hindquarter (HQ), renal (R), and mesenteric (M) vessels of conscious rat.

vs. saline) being reached with the 1-nmol/kg dose (Fig. 4). Hindquarter vascular resistance was dose dependently decreased by the 1- and 3-nmol/kg doses of PAF. The maximum effect ($-56 \pm 7\%$, $P < 0.01$ vs. saline) was achieved by the highest dose (Fig. 4). At no dose or time point was there an increase in hindquarter vascular resistance.

Effect of BN 52021 on regional vascular responses to PAF. A 15-mg/kg dose of BN 52021 had no influence on the resting level of organ blood flow (Fig. 4). Pretreatment with BN 52021 attenuated the increase in mesenteric blood flow and decrease in mesenteric vascular resistance produced by the 0.3-nmol/kg dose of PAF but had no significant effect on the mesenteric vasodilator responses produced by the other doses of PAF (Fig. 4). The mesenteric vasoconstrictor effect of the 3-nmol/kg dose of PAF was significantly attenuated but not completely blocked by BN 52021. The renal vasodilation induced by the 1-nmol/kg dose of PAF was totally blocked by BN 52021 (Fig. 4), and the hindquarter vasodilation produced by the 1- and 3-nmol/kg doses of PAF was significantly attenuated by BN 52021 (Fig. 4).

DISCUSSION

The present report is an extension of our previous efforts to elaborate the mechanisms involved in the cardiovascular actions of PAF in the conscious rat. We have chosen the rat as the experimental model for these studies, since in this species no apparent respiratory effects are associated with the hemodynamic responses to PAF. In addition, at PAF doses used in the present experiment, rat platelets are resistant to PAF-induced aggregation (24), and therefore the hemodynamic effects of PAF in the rat are not confounded by blood cell aggregation commonly seen in other species (e.g., rabbit, pig, guinea pig, and dog).

Intravenously infused PAF decreased blood pressure and increased heart rate in conscious rats. These results are in agreement with the hypotensive responses to PAF described previously in the conscious rat (11, 31) and in a number of other species such as guinea pigs, dogs,

domestic pigs, and rabbits (for review see Ref. 8). The hypotensive effect of the low doses of PAF seems to be due to peripheral vasodilation, since TPRI was markedly decreased at the peak of the hypotensive response while the cardiac index did not change. Opposite to the effect of the low doses of PAF, the 3-nmol/kg dose produced a sustained decrease in cardiac index despite the prominent tachycardic response. At this dose a transient fall followed by sustained elevation of TPRI was also observed. Thus our results are in accordance with the cardiodepressant and vasoconstrictor responses to high systemic doses of PAF in the anesthetized dog (27, 30) and in the pentobarbital sodium-anesthetized rat (12). In vitro studies also indicated a cardiac depressant action of PAF at very high concentrations (5, 18, 26). On the isolated human papillary muscle, PAF produced a transient increase of contractility followed by a prolonged depression (1). In the intact domestic pig, intracoronary infusion of PAF first dilated and then constricted the coronary arteries (10).

In the present study systemic injections of PAF dose dependently decreased renal and hindquarter vascular resistance and in the mesenteric region induced a biphasic response: a vasodilation in the low doses followed by a constriction by higher doses. The regional hemodynamic effects of PAF resemble those obtained by Faber et al. (7), who found hypotensive, tachycardic, and renal, mesenteric, and hindquarter vasodilator responses after intravenous (50–500 ng/rat) injection of the antihypertensive polar renomedullary lipids (APRL) in the conscious rat. However, the regional hemodynamic effects of pure synthetic PAF in conscious rats have not been reported previously. In the anesthetized spontaneously hypertensive rat (SHR), decreases in blood flow to brain, heart, kidneys, lungs, and spleen accompanied the systemic hypotension induced by continuous intravenous infusion of high doses of PAF ($0.3–1$ $\mu g \cdot kg^{-1} \cdot min^{-1}$ equaling $0.6–2$ $nmol \cdot kg^{-1} \cdot min^{-1}$), but the changes in vascular resistance were not reported (12). In a more recent study, 0.06 nmol/kg PAF produced mesenteric vasodilation in anesthetized SHR (19). In the dog, PAF produced renal vasoconstriction (2, 30) but vasodilation when infused into the femoral artery (27). Direct injection of PAF into the coronary circulation of anesthetized domestic pig produced a biphasic change in coronary blood flow: vasodilation followed by vasoconstriction (9).

The mesenteric vasodilation was observed at PAF doses that had no effect on systemic arterial pressure or organ blood flow in the renal or hindquarter regions. Analogous to our findings, the mesenteric artery seemed to be the most sensitive organ for the vasodilator effect of APRL in the rat (7). Dose-dependent mesenteric vasodilation after injections of the PAF precursor 1-palmityl-2-acetyl-glycerol was reported in anesthetized SHR (19). In the present study, the mesenteric vasculature responded to PAF with a biphasic response: after the initial vasodilation a vasoconstrictor response was found at high doses of PAF. In a recent study in the rat, high doses of PAF (2 μg/rat equaling $\sim$10 nmol/kg) induced bowel necrosis (15). Therefore, our finding suggests that the pathological changes found in the intestine
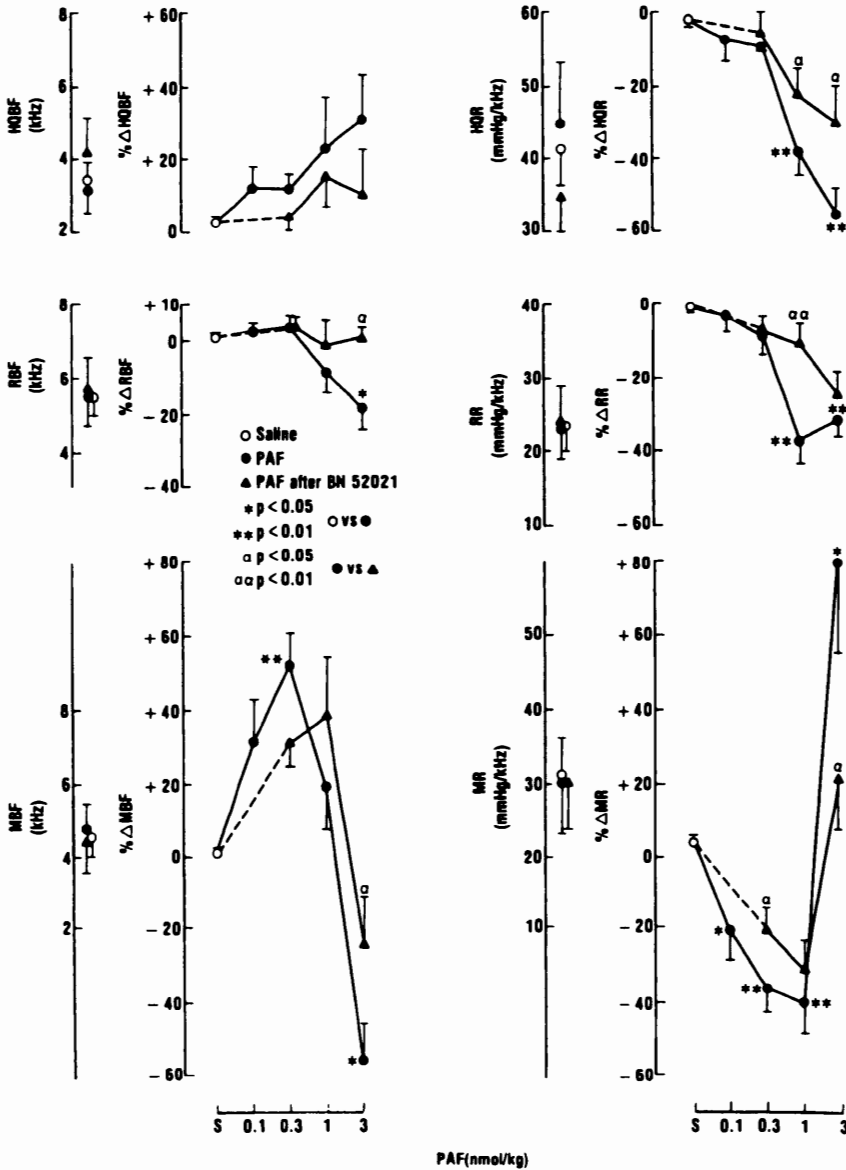
FIG. 4. Effect of 0.1–3 nmol/kg PAF on regional blood flow and vascular resistance in conscious rats. *Left*: each graph denotes base-line levels of blood flow or vascular resistance immediately before PAF administration; *right*: maximum changes 30 s after PAF. BN 52021 was injected intravenously 20 min before PAF administration. Values indicate means ± SE. Asterisks and α denote statistical significance by Student-Newman-Keuls test.

(15) are at least in part the result of ischemia due to a secondary mesenteric vasoconstriction. Furthermore, the increase in mesenteric vascular resistance and reduction of mesenteric blood flow produced by high doses of PAF closely resembles the changes in mesenteric blood flow in acute systemic anaphylaxis (32). Thus ovalbumin challenge in the presensitized conscious rat results in prolonged decrease in mesenteric blood flow even after recovery from the early hypotensive phase (32), indicating that vasoconstriction is maintained. Because PAF is known to be released in the systemic circulation during acute anaphylaxis, systemic lupus erythematosus, and intravascular coagulation in humans (5), our data further contribute to the possibility of PAF-mediated damage to splanchnic blood flow in situations in which large amounts of PAF would be rapidly released.

The PAF antagonist BN 52021 effectively blocked the hypotensive, tachycardic, and cardiac depressant effects of PAF as well as the decrease in TPRI produced by the 1 nmol/kg dose of PAF. We have recently reported (10)

that 5 mg/kg BN 52021 also totally blocked the increase in pulmonary arterial pressure and vascular resistance induced by PAF administration in the neonatal pig. BN 52021 both prevented and reversed the hypotension, hemoconcentration, and plasma extravasation produced by PAF in the rat (24) and antagonized PAF-induced bronchoconstriction in the guinea pig (4). Because BN 52021 is known to be a specific antagonist of PAF receptors (4, 24), our results indicate a direct stimulation of the PAF receptors as a main mechanism of its hypotensive and cardiodepressant actions. In an earlier study, specific cholinergic, histaminergic, or β-adrenoceptor antagonists had no blocking capacity against the hypotensive effect of PAF in the rat (17), whereas in the anesthetized dog or domestic pig the hypotensive effect of PAF is prevented by indomethacin (8, 27). The tachycardic effect of PAF, on the other hand, is blocked by the β-adrenoceptor blocker propranolol (31) and is probably mediated by a reflex activation of the sympathetoadrenomedullary system due to the systemic hypoten-

sion (11, 20). Thus in the pithed rat, which totally lacks the central nervous and spinal autonomic control, PAF produced hypotension with no effect on heart rate or plasma catecholamines (11). Therefore, the attenuation of the tachycardic effect of PAF by BN 52021 in our study can be explained by the blockade of PAF-induced hypotension rather than by a direct inhibition of cardiac PAF receptors.

In the present study only the 3 nmol/kg dose of PAF decreased cardiac output, whereas the lower doses had no consistent effect on cardiac index per se; half of the animals reacted to the 1-nmol/kg dose with a transient increase in cardiac index, whereas in the remaining animals PAF decreased cardiac index. BN 52021 totally blocked the cardiac depressant effect of the high dose of PAF, even though the hypotensive response was only partially attenuated. PAF receptor blockade with BN 52021 or SDZ 63-441 also modified the effect of the 1-nmol/kg dose of PAF so that, after PAF receptor blockade, this dose of PAF induced solely a sustained increase in cardiac index in each animal studied. BN 52021 and SDZ 63-441 had no intrinsic activity of their own on the cardiovascular system and both of these compounds are selective antagonists of the PAF receptors, at the doses used in the present study (4, 13, 19, 24). Therefore, the increase in cardiac index after BN 52021 or SDZ 63-441 treatment is likely to be due to an interaction with the PAF receptors. Unmasking of a positive inotrophic action of PAF by the receptor antagonists could insinuate the existence of two different subclasses of PAF receptors that mediate opposite effects on the cardiac function. This might explain the variable responses of PAF on cardiac function in in vitro studies (1, 11, 18, 26).

PAF receptor blockade by BN 52021 had a differential degree of antagonism against the regional hemodynamic effects of PAF, yet the hindquarter effects were most effectively blocked by BN 52021 with only a partial attenuation of the renal and especially the mesenteric responses. Evidence for a direct action of PAF on blood vessels has also been shown in the dog in which the renal vasoconstrictor effect of intrarenally infused PAF was not affected by blockers on α-adrenergic or angiotensin II receptors but was enhanced by cycloxygenase inhibitors (2). The coronary constrictor effect of PAF in the pig was blocked by indomethacin and thus probably mediated by cycloxygenase metabolites of arachidonic acid, but the coronary dilation was unaffected by cycloxygenase inhibitors (10). Further evidence in support for arachidonic acid metabolites as mediators of the vasoconstrictor effects of PAF was recently provided by Stewart and Piper (26), who found attenuation of the coronary constrictor effect of PAF in the isolated rat heart by cycloxygenase and lipoxygenase inhibitors. The potential role of the arachidonic acid metabolites in the regional hemodynamic responses to PAF in the conscious rat remains to be further studied. However, in the conscious rat, PAF produced predominantly vasodilator responses in all vascular beds despite the potential generation of vasoconstrictor mediators such as thromboxane A_2 and leukotrienes (11, 26). However, indomethacin did not block the hypotension produced by PAF in the rat (11).

In contrast to the recent in vitro findings, our study failed to show any PAF-antagonistic effect of alprazolam. This triazolobenzodiazepine was shown to block PAF-induced aggregation of human, guinea pig, and canine platelets (2, 6, 16). The renal vasoconstrictor effect of PAF in the anesthetized dog was attenuated by alprazolam and by the serotonin blocker methysergide (2). In the conscious rat, however, alprazolam at a relatively high dose (3 mg/kg iv), which already exerted typical intrinsic activity of a sedative (e.g., hypotension), had no effect on the cardiovascular responses to PAF. Species differences might account for the discrepancy between our finding and the previous study on dogs in which serotonin release from platelets might contribute to the PAF actions (2). Because most of the previous reports using alprazolam have been about platelets (2, 6, 16), and rat platelets are known to be resistant to PAF (24), the lack of an antagonistic effect of alprazolam in the rat is not unexpected.

In summary, the results indicate that the systemic hemodynamic changes induced by PAF are due to a combination of cardiac and vascular derangements. PAF is a potent vasodilator of mesenteric > hindquarter = renal vessels at low doses and a cardiac depressant at high doses. The PAF antagonist BN 52021 and SDZ 63-441 effectively blocked the cardiac depressant effect of PAF, and BN 52021 attenuated, in part, the regional vascular responses (hindquarter > renal ≫ mesenteric). Thus specific PAF-antagonists such as BN 52021 or SDZ 63-441 might be used as therapeutic agents in disease states such as endotoxemia, anaphylaxis, and other immune reactions in which increased release of PAF is a contributing factor to key hemodynamic derangements.

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