THE CARDIOVASCULAR AND PLATELET EFFECTS OF EPOPROSTENOL (PROSTACYLIN, PGI₂) ARE UNAFFECTED BY β -ADRENOCEPTOR BLOCKADE IN MAN

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1 Atenolol 0.2 mg/kg i.v., propranolol 0.2 mg/kg i.v. or placebo were given in a double-blind crossover study to six healthy male subjects, and the effects of a subsequent infusion of epoprostenol (prostacyclin, PGI₂) 0–6 ng kg⁻¹ min⁻¹ monitored.

2 PGI₂ caused a tachycardia, a fall in diastolic blood pressure, a rise in pulse pressure, reduction in pre-ejection period (PEP) and rise in left ventricular ejection time index (LVETI), headache and facial flushing at doses of PGI₂ greater than 2 ng kg⁻¹ min⁻¹, ($P < 0.05$).

3 β -adrenoceptor blockade did not prevent the tachycardia in response to PGI₂, and did not interact with any of the other dynamic effects of PGI₂.

4 *In vitro*, PGI₂ at 1 and 2 ng/ml inhibited platelet aggregation to ADP ($P < 0.01$), although no significant effect on platelet aggregation was seen in the *in vivo* study. Atenolol and propranolol at a final concentration of 1 μ g/ml did not affect this *in vitro* effect of PGI₂ on platelet aggregation.

5 Pretreatment with atropine 0.04 mg/kg i.v. in three subjects did not attenuate the tachycardia caused by PGI₂ infusion, even though the baseline heart rate was increased.

6 Adverse effects to PGI₂ infusion included sudden bradycardia, pallor and sweating, suggesting that the Bezold-Jarisch reflex seen in animals in response to PGI₂ may also occur in humans.

7 Neither increased sympathetic drive nor vagal withdrawal are likely causes of the tachycardia following PGI₂ infusion.

Introduction

Epoprostenol (prostacyclin, PGI₂) is synthesised predominantly in the vascular endothelium (Moncada *et al.*, 1977a; Armstrong *et al.*, 1978a). It is a potent inhibitor of platelet aggregation (Gryglewski *et al.*, 1976; Moncada *et al.*, 1977b), a vasodilator (Szczechlik *et al.*, 1980; Eklund *et al.*, 1981) and may be an important endogenous mediator of peripheral vascular resistance (Armstrong *et al.*, 1978b).

In animals, reports on the cardiovascular responses to exogenous epoprostenol are contradictory and it is possible that the action depends on the species studied as well as on the dose of epoprostenol administered (Lefer *et al.*, 1978; Karmazyn *et al.*, 1978). In anaesthetized rat and rabbit i.v. epoprostenol produces hypotension and tachycardia (Armstrong *et al.*, 1978b) while in dogs epoprostenol usually produces bradycardia and hypotension (Chapple *et al.*, 1980;

Chiba & Malik, 1980). However, epoprostenol in dogs produces tachycardia when the dose is low, after atropine pretreatment or after vagotomy (Chiba & Malik, 1980). The precise mechanism(s) underlying these changes are not yet clear.

Epoprostenol infusion in healthy human volunteers produces a dose-related increase in heart rate and a decrease in diastolic blood pressure and pre-ejection period (Warrington *et al.*, 1980). A reflex increase in chronotropic stimulation of the heart secondary to arteriolar vasodilation was the proposed mechanism (Warrington *et al.*, 1980). Others (Fitzgerald *et al.*, 1979) claimed that the increase in heart rate during epoprostenol infusion is a baroreflex in response to the fall in blood pressure and is mediated via vagal withdrawal.

In animals, the tachycardia following epoprostenol

was also thought to be of reflex origin, as the tachycardia could be suppressed by propranolol in dogs (Chiba & Malik, 1980) and was reduced in rats when pithed or following ganglion blockade (Armstrong *et al.*, 1978b). However, in a rat isolated atrial preparation epoprostenol was found to be a powerful inotropic and chronotropic stimulator (Borda *et al.*, 1979). This action of epoprostenol was mediated via the release of tissue catecholamines and was inhibited by propranolol (Borda *et al.*, 1980).

The present study was designed to examine the cardiovascular and platelet effects of exogenous epoprostenol in man following β -adrenoceptor blockade and also the cardiovascular effects of epoprostenol following parasympathetic blockade in order to clarify the roles of increased sympathetic tone and vagal withdrawal in the responses to epoprostenol in man. A preliminary account of this work has already been presented (Fish *et al.*, 1982).

Methods

Materials

Epoprostenol synthesised by Upjohn was formulated by the Wellcome Foundation Ltd, as a sterile freeze dried formulation in vials containing 0.5 mg, and stored at 5°C until use. Immediately before use, epoprostenol was reconstituted with sterile glycine buffer (Wellcome Foundation Ltd) at pH 10.5. Adenosine diphosphate (ADP) was supplied by the Sigma Chemical Company and reconstituted in distilled water to give final concentrations of 0.5, 1, 2, 4, 8, 16, 32 μM , and stored frozen until use. Atenolol and propranolol for human use were supplied by I.C.I. Atenolol and propranolol powder for the *in vitro* studies were supplied by I.C.I. and reconstituted in distilled water to give a final concentration of 1 $\mu\text{g}/\text{ml}$.

Subjects

Healthy, non smoking subjects aged between 20 and 45 years and weighing 60 to 83 kg, participated as follows;

1. *In vivo* β -adrenoceptor blocker-epoprostenol trial, six males with no detectable abnormality on clinical examination or ECG.
2. *In vitro* β -adrenoceptor blocker-epoprostenol trial, thirteen males and females.
3. Atropine-epoprostenol trial, three male subjects.

No medications were taken for 2 weeks prior to studies. The nature, purpose and possible hazards were explained to the subjects and written informed consent obtained.

1. *In vivo* β -adrenoceptor blocker-epoprostenol trial (Figure 1)

The subjects attended the laboratory after an overnight fast. They rested supine on a couch and an i.v. infusion of normal saline was established in a right forearm vein through an Angiocath (18 G) using a Braun Perfusor-E infusion pump. An indwelling cannula was inserted in a left forearm vein for blood sampling. A sphygmomanometer cuff was applied to the left arm. ECG leads were placed on the limbs, a phonocardiograph transducer on the left second intercostal space and carotid pulsation recorded by Elema-Schonander EMT 510 C transducer.

After 30 min rest, the following measurements were recorded:

1. Arterial blood pressure, with a London School of Hygiene Sphygmomanometer, diastolic BP being taken as the point of disappearance of the Korotkoff sounds (Phase 5).
2. Systolic time intervals performed from the simultaneous recording of ECG Lead II, phonocardiogram and the carotid pulse curve (Weissler *et al.*, 1961).
3. Headache on 10 cm visual analogue scale rated by the subject reading from 'no headache' to 'the worst headache I could possibly imagine'.
4. Colour photographs of the subject's face, using a 35 mm SLR camera with Kodachrome 25 film, and constant subject to camera distance. The photographs were later rated for flushing by an independent observer who was blind to the treatment and PGI₂ dose.
5. Platelet aggregatory responses to ADP. Blood samples (9 ml) were collected in plastic tubes containing 1 ml of 3.8% trisodium-citrate. Platelet rich plasma (PRP) was prepared as the supernatant resulting from centrifugation of the blood sample at 120 g for 5 min, using a Petalfuge centrifuge (F. T. Scientific Instruments). After the PRP had been removed by a plastic pipette, the remainder was respun at 600 g for 15 min to obtain the platelet poor plasma (PPP). A solution of ADP (10 μl) was added to a 500 μl aliquot of PRP. Platelet aggregation was measured within 15 min of blood sampling using a Payton Dual Channel aggregometer in response to ADP concentrations ranging between 0.5 μM and 32.0 μM (final concentration).

Following the above measurements, an i.v. injection of atenolol 0.2 mg/kg, propranolol 0.2 mg/kg or normal saline was given (double-blind, and in a randomized order) over 5 min into the right forearm vein, and the above measurements (1–5) were repeated 1 h after injection. The saline infusion was then discontinued and epoprostenol in glycine buffer was infused over consecutive 15 min periods at 0, 2, 4 and 6 ng kg⁻¹ min⁻¹. In the 10th–15th min of each infusion period, measurements (1–5) were repeated.

The degree of β -adrenoceptor blockade was assessed

Figure 1 Experimental protocol for the *in vivo* β -adrenoceptor study.

sed by a bicycle ergometer exercise test 3.5 h after the injection at a load pre-determined for each subject to produce an increase in heart rate to 160 beats/min in the control state.

2. Atropine-epoprostenol study

After an overnight fast, the subjects attended the laboratory, rested supine and an i.v. infusion of normal saline was established as before. Arterial BP and pulse rates were measured. Atropine 0.04 mg/kg was given i.v. over 5 min after 30 min rest. After a few minutes epoprostenol was then infused as before; the arterial BP was measured every 10 min and the pulse rate at least every 5 min.

3. In vitro β -adrenoceptor blocker-epoprostenol study

In this study platelet aggregatory responses to ADP were measured with and without addition of epoprostenol and β -adrenoceptor blocking drugs. Blood (45 ml) was collected by direct venepuncture and delivered into a plastic tube containing 5 ml of 3.8% trisodium citrate, and PRP/PPP prepared as before. The PRP was adjusted with PPP (if necessary) to achieve a platelet count between $250\text{--}350 \times 10^9/l$, platelet counts as measured using a Coulter thrombocounter Model C. Platelet aggregation was measured as before. Dose response curves to 0.5, 1, 2, 4, 8, μM ADP were obtained in the following conditions.

1. Baseline (PRP without any drug)
2. Epoprostenol: 10 μl of epoprostenol solution was added to 500 μl of PRP to give final concentration of 0.5, 1 and 2 ng epoprostenol/ml
3. β -adrenoceptor blocker: 10 μl of either atenolol or

propranolol, was added to 500 μl of PRP to give a final concentration of 1 $\mu g/ml$.

4. β -adrenoceptor blocker and epoprostenol: 10 μl of either atenolol or propranolol (1 $\mu g/ml$ final concentration) was added to 500 μl of PRP followed by 10 μl epoprostenol as in (2).

ED₅₀s of ADP, that is the ADP concentration required to produce 50% of maximal aggregation, were then calculated for 1–4 above.

Statistical analysis

Analysis of covariance was performed to examine the effect of β -adrenoceptor blocking drugs before dosing with epoprostenol. The effect of β -adrenoceptor blocking drugs and epoprostenol was analysed by a split-plot analysis of variance on all the values with subject, epoprostenol dose and β -adrenoceptor blocker as factors. Placebo, atenolol and propranolol were compared using the Newman-Keuls multiple range test. The various doses of epoprostenol were compared with 1 h values using Dunnett's test for the comparison of several treatments with a control. A log transformation was carried out on the scores of headache. Flushing of the face was analysed using Friedman's test. For platelet aggregation data, analysis of variance was performed on the log ED₅₀s.

Results

1. In vivo β -adrenoceptor blocker-epoprostenol study

a Effect of β -adrenoceptor blocking drugs After 1 h and averaged over all times both β -adrenoceptor

blockers produced a drop in the heart rate (P close to 5% significance), a rise in PR interval ($P < 0.01$) and an increase in T-wave amplitude ($P < 0.05$) compared with placebo. Atenolol gave a significant rise in QS_2I (total electro-mechanical systole index) compared with placebo ($P < 0.05$). The mean exercise heart rate showed a significant ($P < 0.05$) drop after atenolol (120.2 beats/min) compared with placebo (149.3 beats/min) but the reduction after propranolol (137.7 beats/min) was not statistically significant.

b *Effect of epoprostenol infusion* (Table 1 and Figure 2). There was a dose-related increase in heart rate with epoprostenol infusion following placebo and

both β -adrenoceptor blockers. The increase was significant ($P < 0.05$) with 6 ng epoprostenol $kg^{-1} min^{-1}$ after placebo and with 4–6 ng epoprostenol $kg^{-1} min^{-1}$ after both β -adrenoceptor blockers ($P < 0.01$) (Figure 2). The systolic BP did not show a consistent change. The diastolic BP tended to drop with epoprostenol infusion; the resultant increased pulse pressure showed a statistically significant change with 6 ng epoprostenol $kg^{-1} min^{-1}$ after placebo and after atenolol ($P < 0.01$). Pre-ejection period showed a significant dose related decrease with epoprostenol infusion following placebo and both β -adrenoceptor blocking drugs while the left ventricular ejection time index (LVETI) showed

Table 1 Cardiovascular and headache response before and during infusion of glycine buffer and epoprostenol following pretreatment with placebo, atenolol or propranolol.

	Treatment	After 1 h	Buffer	PGI_2 (ng $kg^{-1} min^{-1}$)			s.e. difference
				2	4	6	
Heart rate (beats/min)	Placebo	58.5	55.6	59.8	65.2	67.4*	3.16
	Atenolol	50.2	50.3	53.0	58.6**	58.6**	2.38
	Propranolol	48.8	49.3	51.5	60.1**	62.2**	2.49
Systolic BP (mmHg)	Placebo	111.8	114.2	114.5	113.2	118.7*	2.2
	Atenolol	112.8	112.3	113.7	111.8	118.5	5.4
	Propranolol	108.3	108.2	107.7	111.7	114.2	3.0
Diastolic BP (mmHg)	Placebo	71.8	70.3	71.2	65.7	65.5	3.68
	Atenolol	76.0	75.5	75.5	69.8	64.7	7.27
	Propranolol	72.2	72.0	74.3	70.2	67.0	2.52
Pulse pressure (mmHg)	Placebo	40.0	43.8	43.3	47.5	53.2**	3.55
	Atenolol	36.8	36.8	38.2	42.0	53.8**	4.35
	Propranolol	37.7	36.2	33.3	41.5	47.2	4.08
PEP (ms)	Placebo	123.0	123.3	117.2*	108.8**	102.5**	1.94
	Atenolol	136.8	135.5	130.5	120.2**	118.3**	3.19
	Propranolol	130.2	130.2	124.2*	114.2**	109.2**	2.28
QS_2I (ms)	Placebo	491.3	493.3	489.2	485.0	486.2	3.2
	Atenolol	501.5	500.8	515.0	499.0	500.2	10.35
	Propranolol	495.2	496.5	494.2	492.0	491.8	1.88
LVET I (ms)	Placebo	362.7	364.8	366.0	369.7	377.2**	3.30
	Atenolol	359.5	360.5	362.3	373.3**	376.0**	2.39
	Propranolol	360.3	361.8	364.5	371.8**	376.3**	1.99
T-wave amplitude (mm)	Placebo	7.9	8.6	7.8	7.4	7.0*	0.31
	Atenolol	11.5	11.7	11.2	10.4*	10.1**	0.34
	Propranolol	10.8	10.9	10.7	9.6**	9.2**	0.29
Headache score (Log)	Placebo	0.76	0.76	0.85	1.29**	1.66**	0.14
	Atenolol	0.8	0.85	1.0	1.35**	1.69**	0.18
	Propranolol	0.8	0.85	0.94	1.16*	1.66**	0.12
Flushing score (ranks)	Placebo		1.4	2.0	3.4	3.2	
	Atenolol		1.2	2.0	2.8	4.0	
	Propranolol		1.8	2.5	2.5	3.3	

PEP pre-ejection period

LVET I Left ventricular ejection time index

QS_2I Total electro-mechanical systole index

* significantly different from 1 h value $P < 0.05$

** significantly different from 1 h value $P < 0.01$

Figure 2 The effect of PGI₂ infusions preceded by placebo (■) or β -adrenoceptor blocking drugs (○ propranolol, ● atenolol) on the heart rate. * $P < 0.05$, ** $P < 0.01$.

increase which was significant with 6 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following placebo and with 4–6 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following both β -adrenoceptor blocking drugs ($P < 0.01$). The QS₂I was unchanged by epoprostenol infusion. The T-wave amplitude decreased significantly with 6 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following placebo ($P < 0.05$) and with 4–6 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following both β -adrenoceptor blockers ($P < 0.01$). The PR interval, QRS duration and QT interval did not show a significant change during epoprostenol infusion.

Headache scores were higher ($P < 0.01$) with 4–6 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following placebo and both β -adrenoceptor blockers. Flushing increased significantly ($P < 0.05$) with epoprostenol infusion after placebo and atenolol, but not after propranolol.

c Effect of epoprostenol infusion on platelet aggregation measured ex vivo Neither β -adrenoceptor blockers showed a significant effect on platelet aggregation

1 h after administration (Table 2). When epoprostenol was infused, the ADP dose ratios were higher though not significantly so except with 4 ng epoprostenol $\text{kg}^{-1} \text{min}^{-1}$ following atenolol.

d Adverse effects to epoprostenol Flushing of the face, a subjective feeling of warmth on the face and headache were almost universally found with epoprostenol. Most subjects experienced these effects with 4 ng $\text{kg}^{-1} \text{min}^{-1}$ PGI₂, although some required a higher dose. Erythema over the vein into which PGI₂ was being infused was seen on several occasions, and with the other adverse effects is discussed in more detail elsewhere (Pickles & O'Grady, 1982).

A more alarming adverse effect was seen on five occasions in three subjects, one subject experienced it every time he received PGI₂. The usual tachycardia in response to PGI₂ rapidly reverted to bradycardia and was accompanied by pallor, sweating and nausea. One subject experienced this just after the infusion had been discontinued, but in the others the infusion was stopped and recovery took place over about 10

Table 2 Mean log platelet aggregation dose-ratio increase from baseline, before and during infusion of PGI₂, and following pretreatment with placebo or β -adrenoceptor blocking drug (*in vivo* study).

Treatment	PGI ₂ (ng $\text{kg}^{-1} \text{min}^{-1}$)				
	After 1 h	Buffer	2	4	6
Placebo	0.19	0.12	0.21	0.2	0.28
Atenolol	0.26	0.23	0.25	0.36	0.48
Propranolol	0.35	0.38	0.37	0.46	0.56

min. This effect occurred only at the higher doses of PGI_2 , and the pallor and bradycardia in these few subjects then disguised the true degree of tachycardia and flushing seen with PGI_2 (Table 1, Figure 2).

2 Atropine-epoprostenol trial

In each of the three subjects atropine produced the expected anticholinergic response, with an increase in heart rate, tachycardia, dry mouth, mydriasis and paralysis of accommodation. The subsequent epoprostenol infusion, however, produced the same further increase in heart rate as it had done on a previous occasion without atropine (Figure 3).

3 Effect of β -adrenoceptor blockers and epoprostenol on platelet aggregation measured in vitro

No significant effect was found with either atenolol or propranolol. With epoprostenol 1 ng/ml and 2 ng/ml the ED_{50} of ADP increased significantly ($P < 0.01$) (Table 3). No significant interaction between epoprostenol and either β -adrenoceptor blocker was found.

Discussion

An increase in heart rate has been reported whenever epoprostenol infusions have been given to human subjects. In the present study, the tachycardia has been modest, about 10 beats/min following 6 ng

$\text{kg}^{-1} \text{min}^{-1}$, but this effect was significant and dose-related. As such an increase in heart rate could be deleterious for some patients who might require the therapeutic use of epoprostenol, we have attempted to determine the mechanism of the tachycardia, and to see if β -adrenoceptor blockade could be used to protect such patients.

While an increase in heart rate during epoprostenol infusion is the usual response in man, in animals the response is more variable. The rat and rabbit respond to epoprostenol with an increase in heart rate (Armstrong *et al.*, 1978 a,b) which is thought to be of reflex origin. In the cat epoprostenol causes a modest tachycardia followed by bradycardia (Lefer *et al.*, 1978). Epoprostenol in 'small' doses ($< 0.1 \mu\text{g/kg}$) produced a fall in BP and tachycardia in dogs (Chiba & Malik, 1980) while larger doses produced a more profound hypotension that was associated with marked bradycardia (Chapple *et al.*, 1980; Chiba & Malik, 1980).

Epoprostenol has been shown to be a vasodilator in man and in animals (Szczeklik *et al.*, 1980; Chapple *et al.*, 1980) and a fall in diastolic blood pressure has been reported here as in other studies. The most logical explanation for the tachycardia following epoprostenol, then, is a sympathetically-mediated reflex in response to this hypotension. Whenever tachycardia is seen with epoprostenol in animals, it can be reduced by the β -adrenoceptor blocker propranolol (Chiba & Malik, 1980) presumably by antagonizing sympathetic drive. However, in our experiments neither the cardioselective β -adreno-

Figure 3 The effect of PGI_2 infusion alone and when preceded by atropine on the heart rate (single subject).

Table 3 Platelet aggregatory responses to ADP and the effect of atenolol and propranolol (1 μ g/ml) and PGI₂ (0.5–2 ng/ml). ADP ED₅₀s (*in vitro* study).

	PGI ₂ (ng/ml)				
	Baseline	0.5	1.0	2.0	
Atenolol n = 6	2.20	2.60	3.64	4.73	
Control n = 6	2.17	2.87	3.77	4.83	
Propranolol n = 7	2.82	3.57	3.77	5.08	
Control n = 7	2.61	3.56	3.95	5.10	
Overall means	2.47	3.18	3.79**	4.95**	s.e. difference 0.368

** Significantly different from baseline value $P < 0.01$

ceptor blocker atenolol, nor the non-selective agent propranolol attenuated the increase in heart rate seen with epoprostenol. Although it could be argued that we used an insufficient dose of β -adrenoceptor blocker, we were able to show reduction in exercise tachycardia. Consistent with our results, we note that Fitzgerald *et al.* (1979) failed to find an increase in plasma noradrenaline accompanying epoprostenol-induced tachycardia, as would have been expected during increased sympathetic activity. It has been argued that the stimulus for reflex tachycardia is a fall in mean blood pressure, and because of the increase in pulse pressure we found no significant change in this variable.

Our experiments appear to exclude increased sympathetic drive as a cause of the tachycardia. Fit young subjects are likely to have considerable resting vagal tone—could withdrawal of this lead to tachycardia with epoprostenol? Although we have performed relatively few experiments in the presence of atropine, we have found no evidence for this mechanism either, with the same increase in heart rate found as in the absence of atropine. It has been proposed that in dogs the heart rate response to epoprostenol depends on the basal heart rate, and tachycardia is seen only if the basal rate is low (Chiavarelli *et al.*, 1982). We have found no correlation of the heart rate response to epoprostenol with basal heart rate in over 40 experiments with basal rates ranging from 45 to 115 (following atropine, β -adrenoceptor blocker or no pretreatment, and including unpublished experiments).

In dogs, the predominant cardiac response to epoprostenol is bradycardia, particularly if the basal heart rate is high, and this appears to be mediated by a vagal reflex (Chapple *et al.*, 1980; Hintze *et al.*, 1979, 1981) involving chemosensitive C-fibres in the

heart (Roberts *et al.* 1980) or lung (Armstrong & Miller, 1980). In several subjects we have noted a sudden bradycardia, nausea, pallor and sweating that is indistinguishable clinically from sudden vagal activity. In view of the relative frequency with which we have seen this, we were surprised to find very few similar previous reports from human studies, (for review, see Pickles & O'Grady, 1982). On one occasion we saw this effect when our subject was incompletely atropinised, and apart from the lack of sweating, the collapse was indistinguishable from those we had seen on previous occasions. It seems highly likely that we have observed the same type of Bezold-Jarisch reflex as described in animals studies, and thus the same mechanisms may be involved in the cardiovascular responses to epoprostenol in humans.

The main mechanism for the tachycardia to epoprostenol in humans is unresolved. There is little evidence that epoprostenol has a direct chronotropic effect (Chiba & Malik, 1980) and that which has been reported is blocked by propranolol (Borda *et al.*, 1979), but more invasive methods than those employed here would be needed to determine this conclusively.

We found that epoprostenol infusions were associated with a significant increase in LVETI; this may reflect an increase in stroke volume (Weissler *et al.*, 1961). An increase in cardiac index with epoprostenol has previously been reported (Bergman *et al.*, 1981). With epoprostenol infusion we found a reduction in T-wave amplitude, and with β -adrenoceptor blockade an increase, suggesting that T-wave amplitude may be inversely related to the heart rate. This effect of β -adrenoceptor blockade on T-wave height has been reported previously (Stern & Eisenberg, 1969). Epoprostenol caused a reduction in PEP: this result is expected if peripheral resistance

or arterial pressure falls and/or a positive inotropic action is exerted (Gibson, 1978).

Epoprostenol infusion failed to show a statistically significant inhibition of platelet aggregation when measured *ex-vivo* probably because of the relatively modest doses administered compared with those producing significant inhibition in other studies. When platelet aggregation was measured *in vitro* after epoprostenol addition, the ED₅₀ of ADP showed a significant increase indicating a significant inhibition of platelet aggregation. This epoprostenol-induced inhibition of platelet aggregation was not modified by the presence of atenolol or propranolol. In our experiments, atenolol and propranolol were without effect on ADP-induced platelet aggregation. Recent reports have suggested that the long-term use of β -adrenoceptor blockers can be associated with the inhibition of platelet aggregation and this might contribute to the cardioprotective effects of these drugs (Campbell *et al.*, 1981). It is possible that this effect may only be demonstrable in patients with arterial

disease and hyperactive platelets (Weksler *et al.*, 1977; Mehta *et al.*, 1978) and the platelets from normal subjects are unaffected by propranolol even when it is given *in vivo* (Leon *et al.*, 1978).

The studies reported here do not lead to firm conclusions regarding the mechanisms involved in the cardiovascular responses to epoprostenol. However, we now know that β -adrenoceptor blockers can be given safely to patients receiving epoprostenol and no attenuation of the action of epoprostenol is to be expected. Based on our experience with a single subject, we could also conclude that atropine is no antidote to the Bezold-Jarisch type reflex that is occasionally seen with epoprostenol, in keeping with the incomplete blocking effect it has on this reflex in animals (Chapple *et al.*, 1980).

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