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## Experience with Prostacyclin in Cardiopulmonary Bypass in Dog and Man

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## Experience with prostacyclin in cardiopulmonary bypass in dog and man

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[Plate 1]

The activation and disruption of platelets resulting from contact with bypass equipment is responsible for bleeding problems after open-heart surgery and for vascular injury leading to cerebral damage. This paper presents experimental and clinical evidence for the benefits of the preservation of platelets by prostacyclin ( $\text{PGI}_2$ ) during cardiopulmonary bypass operations.

Work with dogs showed that  $\text{PGI}_2$  was more effective than heparin in maintaining the numbers and aggregability of the platelet population, but by far the most effective was a combination of the two agents.

A subsequent double-blind clinical trial on 24 patients undergoing coronary vein grafts, in which the combination was compared with heparin alone, confirmed these findings in man. In the presence of  $\text{PGI}_2$ , platelet numbers and aggregability were preserved, with a consequent reduction in blood loss. Significantly fewer reinforcing doses of heparin were required by the  $\text{PGI}_2$  group. The integrity of platelets in the presence of  $\text{PGI}_2$  was reflected by the lack of micro-aggregates and fibrin deposits on arterial line filters. In both the human and dog studies,  $\text{PGI}_2$  was shown, by the cultured foetal mouse heart test, to prevent the release of circulating cardiotoxic factors during bypass.

The known vasodilator effect of  $\text{PGI}_2$  was observed but caused no clinical problems. An unexpected feature was the maintenance of perfusion pressure without the need for additional fluid. This may indicate that  $\text{PGI}_2$  reduces capillary permeability.

## INTRODUCTION

The discovery of prostacyclin ( $\text{PGI}_2$ ) in 1976 (see Moncada & Vane 1978), the most powerful inhibitor of platelet function described so far, was of immediate interest to those of us who are concerned about the unsatisfactory results of contemporary cardiopulmonary bypass procedures. Many of the problems after open-heart surgery are attributable to deficiencies of the haemostatic system. Post-operative bleeding, embolism and thrombosis are occasionally a threat to life and very commonly a cause of post-operative morbidity. Heparin anticoagulation and protamine reversal is still the only technique available for routine use in open-heart surgery. For many years this combination has been recognized as less than satisfactory (Bass & Longmore 1969; Åberg 1974).

The adverse effects of protamine, a family of basic polypeptides, are well known (Larkin *et al.* 1978; Ellison *et al.* 1978; Velders 1980). The use of heparin, a family of complex polysaccharides, while preventing gross coagulation of blood, does not inhibit the first stages of the clotting cascade. Platelets are activated and destroyed as a result of the contact between blood and the artificial surfaces of the heart–lung machine. Heparin may itself exacerbate such destruction (Wolf 1967).

After open-heart surgery, the patient's recovery is always very much slower than after closed-heart or general surgery. Post-operative complications include varying degrees of

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psycho-pathological disturbances. Some patients are unable to work or live in harmony with their families after contemporary open-heart surgery. Surgical life-saving triumphs are all too commonly marred by disabling complications. These complications are more serious than might be expected from the loss of some platelets. Platelet adhesion to the extracorporeal apparatus and aggregation, not prevented by heparin, leads to the release of platelet granular constituents. ADP, 5-hydroxytryptamine and the small proteins  $\beta$ -thromboglobulin ( $\beta$ TG) and platelet factor 4 (PF4) are released into the circulation.

Some of the released substances are procoagulant (Broekman *et al.* 1975), and when caused by abnormal platelet activation may seriously disturb the chemical balance regulating normal endothelial repair in haemostasis. There is evidence supporting the theory that the heparin-neutralizing activity present in blood is due to PF4 released from platelets (Niewiarowski *et al.* 1976).  $\beta$ TG is a significant component of this protein secretion and may inhibit local production of PGI<sub>2</sub> (Hope *et al.* 1979).

During cardiopulmonary bypass, haemodiluting fluids used in the prime and administered to the patient pass rapidly into the extravascular space, suggesting to us that there is increased capillary permeability. We do not know whether this is related to release of 5-hydroxytryptamine and other substances from adherent and disrupting platelets.

#### DOG EXPERIMENTS

We set out in 1978 to establish in the dog whether the use of PGI<sub>2</sub> would eliminate anti-coagulant-related complications of bypass. Initially we undertook three series of experiments (Longmore *et al.* 1979), using PGI<sub>2</sub> alone, a control group with heparin alone and a series with routine heparinization together with PGI<sub>2</sub>.

In all three sets of experiments similar anaesthetic and surgical techniques were used. A simple bypass circuit was used with gravity drainage from an 8 mm atrial basket to a Bentley paediatric Q110 bubble oxygenator. This was connected to a single-roller pulsatile arterial pump with a finely adjustable driven roller to avoid trauma to the blood. A special feature of the bypass circuit was the inclusion of two filters in series in the arterial line. Pressure take-off points proximal to the first filter, between the filters, and distal to the second filter were used to detect clogging of the filters. Scanning electron microscopy of both sides of both filters was used to determine whether material found on the filter surfaces was deposited on or generated by the filters.

#### *Bypass with PGI<sub>2</sub> alone*

With the use of the circuit described above, PGI<sub>2</sub> alone was used for 1 h bypass in 14 beagles. Remarkably, we achieved long-term survival in all but the first in which there was a technical failure unrelated to the use of PGI<sub>2</sub>. During the surgery in this group, the wounds were dry, appearing as they are in conventional operations performed without extracorporeal circulation and heparinization. There was no measurable blood loss during or after the surgery. Nevertheless, the plasma fibrinogen was reduced and there was frank clot in those regions of the extracorporeal circulation in which there was stasis. There were clots on both filters.

At this stage of the development of oxygenators, reservoirs and filters, the designs for heat exchange and debubbling depend on blood standing in contact with heat exchange surfaces and in holding areas. Until a new generation of apparatus, already designed on different principles, becomes acceptable it will probably not be possible to use PGI<sub>2</sub> on its own.

*Heparin alone*

We undertook two further sets of experiments on greyhounds. In the first six control experiments the dogs were given heparin 300 i.u./kg intravenously (i.v.) 5 min before starting a 2 h bypass.

*Heparin plus PGI<sub>2</sub>*

A further eight experiments were undertaken by using the same heparin routine and prostacyclin at 10 ng kg<sup>-1</sup> min<sup>-1</sup> i.v. for 15 min before bypass and then 1 µg kg<sup>-1</sup> min<sup>-1</sup> into the venous return line near to the heart so that the PGI<sub>2</sub> was in maximum concentration in the oxygenator for the 2 h of bypass. All dogs were given protamine sulphate in a 1:1 ratio to the total heparin dose at the end of bypass.

Blood samples were taken after induction of anaesthesia, when the PGI<sub>2</sub> infusion was started, at the beginning of bypass, every 15 min during and after bypass, and after protamine administration until recovery. These samples were used for haematocrit determination, platelet counts (phase contrast microscopy), and testing on foetal hearts. Platelet-rich plasma was obtained by centrifugation of citrated blood at 1000 g. Aggregation was induced by adenosine diphosphate (20–100 µmol/l) and measured as the increase in light transmission over 2 min in a Born-type aggregometer (Payton dual-channel). Total clotting fibrinogen, thrombin-clotting time, as well as the above measurements, were estimated at the same times and 24 h after operation.

Filters were rapidly removed after bypass and fixed for scanning electron microscopy. During the bypass and during wound closure any bleeding was noted. The pressure differential across the two filters in the arterial line was recorded every 5 min during bypass.

To study possible changes in the plasma proteins and any toxic substances that might be released from damaged platelets, foetal mouse hearts in organ culture were used (Wildenthal 1970, 1971 *a, b*; Hughes & Longmore 1972; Longmore & Smith 1980). Foetal hearts were taken from Theier's (T.O.) pure-strain mice that had been mated 15 days previously; 15 control and 15 test hearts were each cultured in 2 ml of 'Wellcome' 199 culture medium and 1 ml of plasma with cortisol (1 µg/ml) and insulin (50 µg/ml). The plasma was obtained from samples of blood drawn from the oxygenator just before the end of bypass. The hearts were cultured in 95% O<sub>2</sub> and 5% CO<sub>2</sub> at 37 °C. They were examined under a dissecting microscope every 24 h and returned to the incubator when the beating rate had been recorded.

*Results of preliminary dog experiments*

The results of the dog experiments were unequivocal. In the series with heparin only the wounds remained wet throughout the operation. There was some post-operative oozing from the chest wound. All the dogs recovered slowly. One died 6 h post-operatively of haemorrhage. Two more died within 24 h. The platelet count, corrected for haemodilution (figure 1), fell to below 50% during the bypass and even further to 35% after protamine was given. The few platelets remaining in the circulating blood after the protamine injection were incapable of aggregation (figure 2). The plasma fibrinogen was reduced. The pressure differential across the filters in three of the dogs showed a fluctuating pattern: building up, falling, and then building up again.

In the protein-denaturation studies on mouse foetal hearts, there was a significant depression of beating-rate and reduction of survival time (compared with the control plasmas) in hearts receiving plasma from dogs bypassed with heparin alone (figure 3). Scanning electron microscopy

scopy showed a build-up of platelet aggregates on both sides of both filters. The platelets covering the surfaces were breaking down and fibrin strands were forming with red and white cells enmeshed (figure 4*a*, plate 1).

In contrast, in the series combining heparin and PGI<sub>2</sub>, there was no fall in platelet count, no change in platelet activity, and no fall in plasma fibrinogen. Furthermore, there was no increase

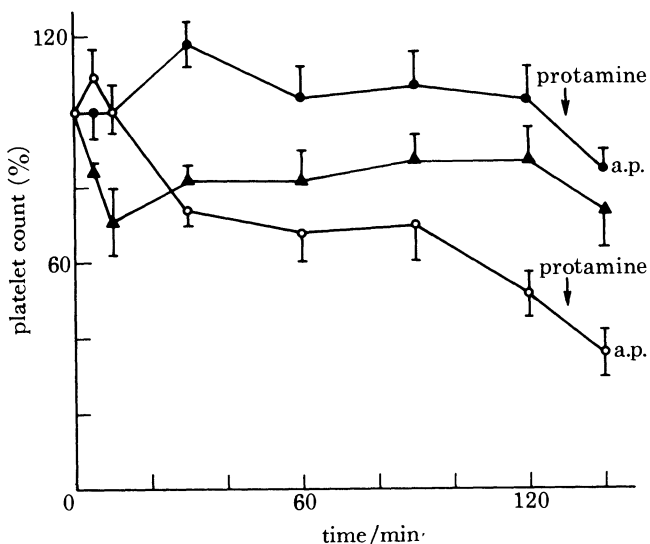


FIGURE 1. Platelet counts during 2 h bypass operations on dogs with heparin alone (○), PGI<sub>2</sub> alone (▲) and a combination of the two (●). A.p., after protamine.

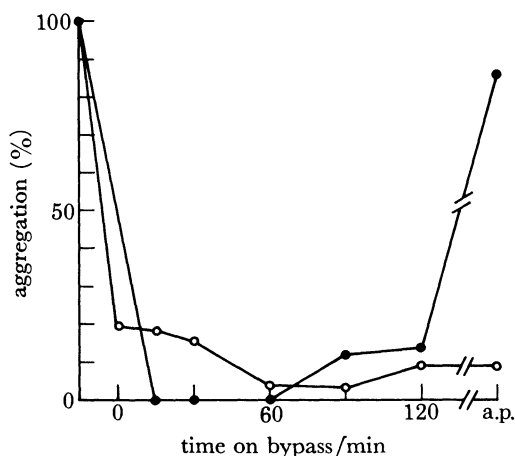


FIGURE 2. Aggregability of dog platelets to ADP during bypass with heparin alone (○) and with heparin plus PGI<sub>2</sub> (●). The ability of platelets to aggregate after a conventional bypass is about 10% of control. This was improved to over 80% with heparin and PGI<sub>2</sub>. A.p., after protamine.

in pressure across the filters during the 2 h of bypass. Platelet aggregation returned to normal within 30 min of the end of prostacyclin infusion. Electron microscopy showed that the filter mesh had only a few adhering platelets. The platelets that had adhered did not appear to have broken down. There were no fibrin strands formed (figure 4*b*). Plasma taken at the end of bypass from these dogs had no significant effect on the beating rate and survival time of the foetal hearts.

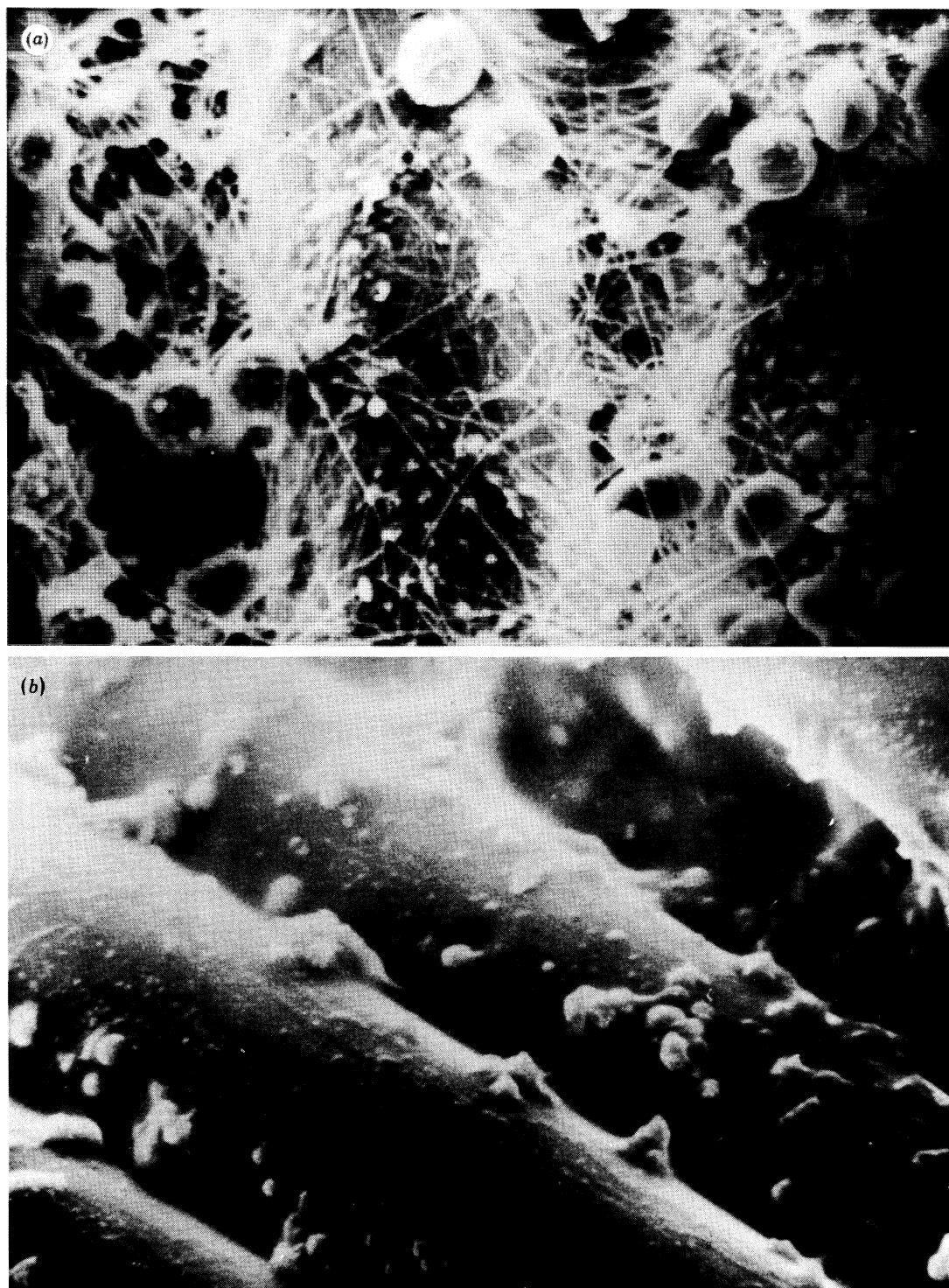


FIGURE 4. (a) Scanning electron microscopy showed a build-up of platelet aggregates, fibrin strands and red and white cells on the filter from the bypass with heparin alone. (b) Scanning electron microscopy showed few platelet aggregates and no fibrin strands when PGI<sub>2</sub> was used in combination with heparin.

(Facing p. 403)

The advantageous effects of prostacyclin in the experiments were not accompanied by any apparent disadvantages. The remarkably low mortality rate in the PGI<sub>2</sub> dog series strongly suggested that the use of PGI<sub>2</sub> in human bypass would be safe. It is well known that the dog is less able than man to withstand the trauma of cardiopulmonary bypass.

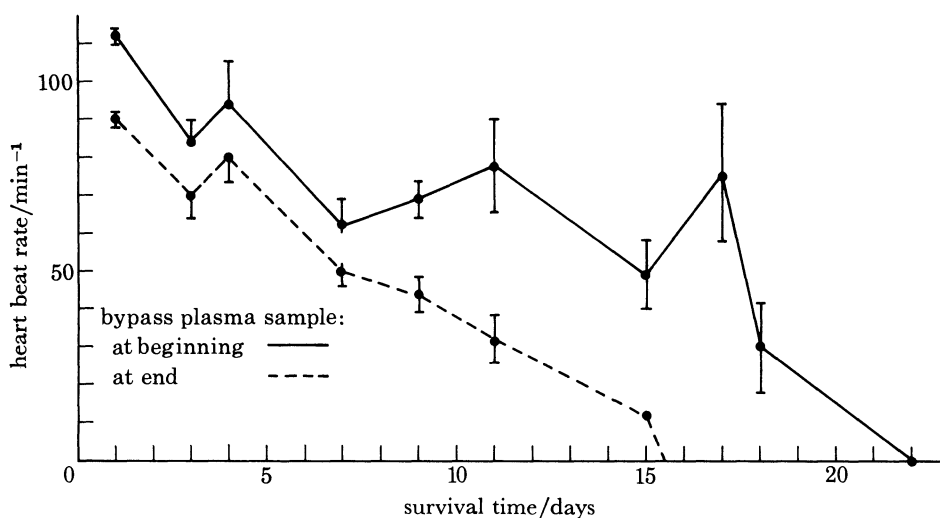


FIGURE 3. Influence of dog plasma on heart rate and survival time of cultured foetal mouse hearts, showing the reduction of these by plasma from the 'heparin only' group. This difference was not seen in the presence of PGI<sub>2</sub>.

#### THE CLINICAL TRIAL

Encouraged by the positive results obtained in the dog laboratory, we undertook a double-blind trial in man, by using PGI<sub>2</sub> in combination with routine heparinization (Longmore *et al.* 1981). This trial was designed to establish whether the beneficial effects of PGI<sub>2</sub> seen in the dog would translate to the human. In addition we also set out to establish whether there would be any measurable clinical benefits from the improved haematological picture obtained when PGI<sub>2</sub> was used. We concentrated on post-operative bleeding and multi-organ damage including brain damage.

#### *Patients and clinical aspects*

The study was carried out on 24 male patients between ages of 36 and 64 years (mean 52 years) in which 12 received PGI<sub>2</sub> and 12 received placebo. One patient (subsequently found to be receiving placebo) was excluded from this study because he had to be returned to theatre with cardiac tamponade. Patients undergoing coronary vein grafts were chosen because they are a homogeneous group operated on with a closed heart. The operating techniques used utilise minimal suction and cause minimal tissue damage. Thus, as in the preliminary study on dogs, most activation of platelets would be due to the traumatic passage of blood through the extracorporeal apparatus. The patients were operated on by Mr Donald Ross, Mr Magdi Yacoub and Mr Graeme Bennett at the National Heart Hospital, using similar techniques. A common anaesthetic protocol was used in every case.

In all the patients a routine bypass circuit with a Bos 10 Bentley oxygenator and Sarn's pumps

was primed with 2 l of Hartmann's solution and 3000 i.u. heparin added. Flow rates varied between 2.2 and 2.4 l m<sup>2</sup> min<sup>-1</sup>. Three suckers were available for the surgeons. These discharged into a Bentley cardiotomy reservoir with a 40 µm in-line filter. No arterial line filtration was used. PGI<sub>2</sub> (synthesized by Upjohn Co. and formulated by the Wellcome Foundation Ltd), reconstituted from the freeze-dried sodium salt (0.5 mg) in glycine buffer (pH 10.5, Wellcome Foundation Ltd) and diluted to the required concentration in normal sterile saline (or placebo (Wellcome Foundation Ltd) containing all ingredients except active drug), was infused by a peristaltic Tekmar drip pump through a central venous line. Infusion of PGI<sub>2</sub> or placebo was started after induction of anaesthesia at a dose rate of 10 ng kg<sup>-1</sup> min<sup>-1</sup> and increased to 20 ng kg<sup>-1</sup> min<sup>-1</sup> at the beginning of bypass. Heparin was given routinely (9000 i.u./m<sup>2</sup> body surface area) after saphenous vein mobilization was completed, and haemostasis was obtained before cannulation of the great vessels. Heparinization was monitored throughout by measuring activated clotting time by means of a Hemochron instrument (International Technidyne Corporation). Reinforcing doses of heparin were given when the Hemochron time fell below 350 s. The PGI<sub>2</sub> or placebo infusion was stopped at the end of bypass and protamine given in a 1:1 ratio to the total heparin dose. Blood and perfusion pressures were recorded continuously and any facial flushing or other unusual features noted. Blood given, blood loss, urine output and fluid given were also recorded before, during and after bypass.

TABLE 1. SAMPLING PROTOCOL

1. During the 24 h pre-operative period (psychometric and neurology tests only)
2. Immediately after induction of anaesthesia to obtain control levels
3. 30 min after beginning of infusion of PGI<sub>2</sub> or placebo
4. 5 min after heparin administration
5. 5 min after start of bypass (when adequate mixing of priming fluid and blood has taken place (foetal heart test only)
- 6a. )
- 6b. ) Every 30 min on bypass
- 6c. )
- 6d. )
7. At the end of bypass before cardioactive drugs are administered (foetal heart test only)
8. 15 min after protamine administration
9. 2 h after bypass
10. 6 h after bypass (CK and BTG assays only)
11. 24 h after bypass
12. 3 days after bypass (CK and BTG assays and psychometric testing).
13. 6 days after bypass (psychometric and neurological tests)

#### *Haematological and biochemical tests*

Blood samples were taken from the radial arterial line or, during bypass, from the arterial reservoir of the pump circuit via a fine catheter.

Sampling times covered the period from induction of anaesthesia to the day after surgery (table 1). Samples were taken at times 2, 3, 4, 6a, 6b, 6c, 8, 9, 10 and 11 for the following haematological tests: haematocrit, haemoglobin, white blood cell count, platelet count, platelet aggregation, prothrombin time, thrombin time, partial thromboplastin time, fibrinogen, anti-thrombin III, and β-thromboglobulin.

β-Thromboglobulin levels were measured by radioimmunoassay with the double antibody (Ludlaim *et al.* 1975).

In addition to these investigations, creatine kinase (including MB isoenzymes) levels were also measured at times 9, 10, 11 and 12.



Post-operative urine samples were collected for creatinine clearance studies. The urine samples were divided into three to avoid loss through accidental spillage or nurses discarding the samples. Platelet aggregability was measured by the Born (1962) technique with a Payton aggregometer. The latter was measured in platelet-rich plasma within 2 min of the blood sample being withdrawn. The degree of aggregability was related to the maximum aggregation attainable with ADP in each specimen.

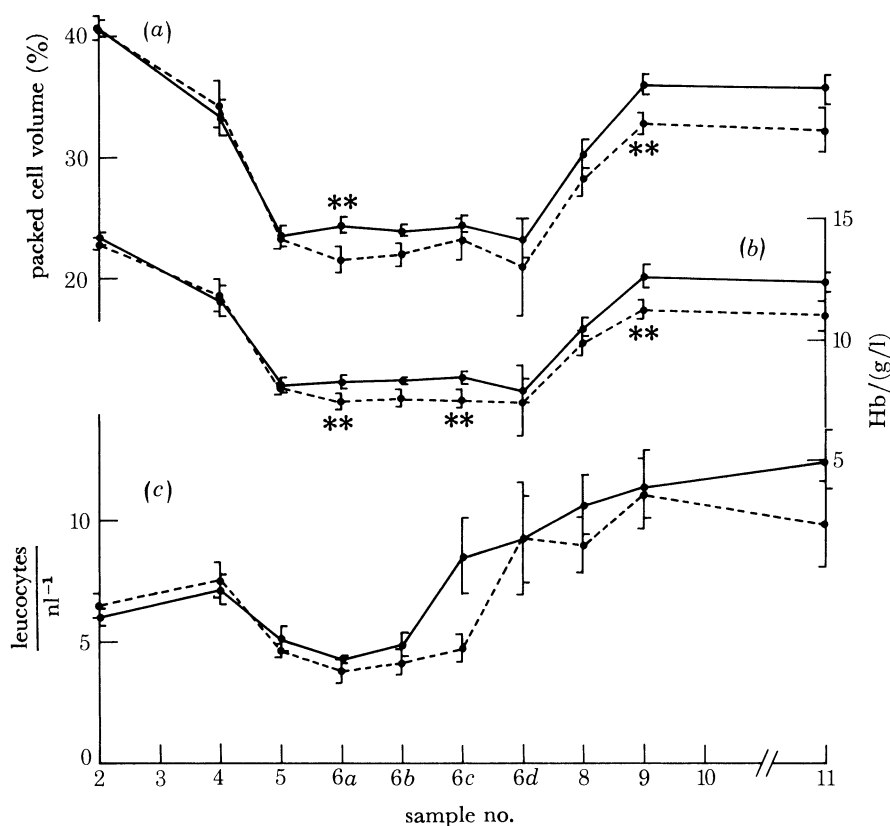


FIGURE 5. Values of haematocrit (a), haemoglobin (b) and white cell count (c) up to 24 h after bypass with (—) and without (---) PGI<sub>2</sub>. These indicate the effect of haemodilution. The steady rise in leucocyte count is to be expected as a result of surgical intervention. \*\*,  $p < 0.05$ ; bars indicate standard error.

The foetal heart test was used as in the dog experiments on samples taken at times 5 and 7.

Psychometric testing was undertaken at times 1, 12 and 13. These tests are designed to detect any post-operative cerebral dysfunction. The testing plan at each testing time (Bethune 1980) included short-term memory recall, the 'Tooting Bec' questionnaire, a digital test, a number connection test and questions relating to the distant past. The complete test takes 20–25 min. At sample times 1 and 13, a routine clinical neurological examination with the use of a fixed protocol, including assessment of cranial nerve function, cerebellar function, muscle power, reflexes and coordination, was undertaken.

## RESULTS

Haemoglobin and haematocrit, but not white cell count, showed slight but occasional significant differences between the placebo and active groups ( $p < 0.05$ ; see figure 5). The results in the PGI<sub>2</sub> group were higher at times during the bypass and consistently higher after bypass. These three variables also indicate the extent of haemodilution at various stages throughout and

after bypass. This indication of dilution can be used to apply a correction factor to other measurements such as platelet counts.

There was no significant difference between the control and test group for prothrombin time, partial thromboplastin time, fibrinogen, antithrombin III, creatine kinase and CK and MB isoenzymes, and  $\beta$ -thromboglobulin.

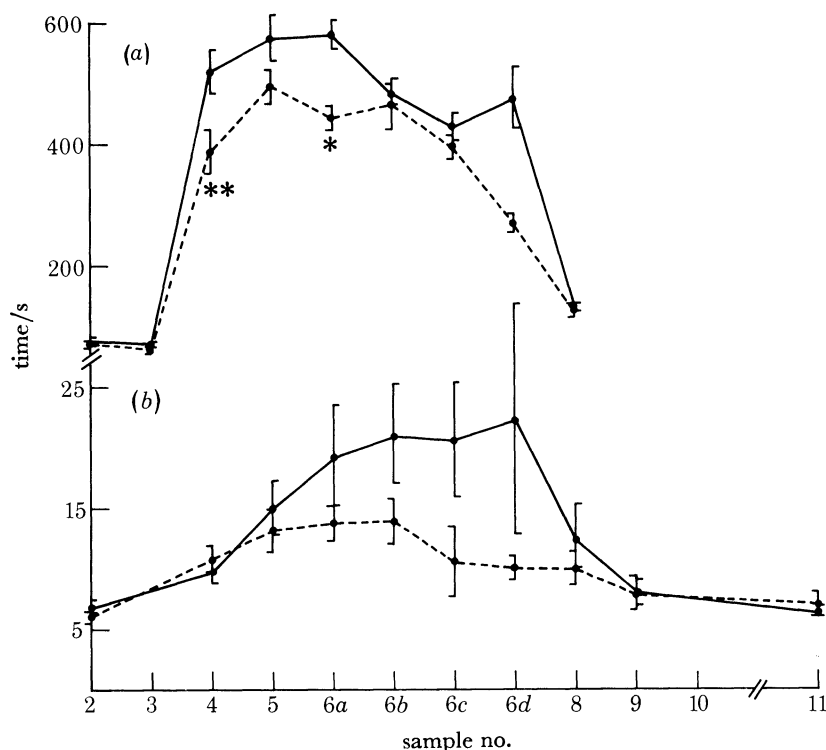


FIGURE 6. The variation of Hemochron value (a) and thrombin time (b) with (—) and without (---) PGI<sub>2</sub>. Both variables were lower in the placebo group throughout bypass in spite of the higher dosage of heparin required by these patients. \*,  $p < 0.01$ ; \*\*,  $p < 0.05$ ; bars indicate standard error.

In the PGI<sub>2</sub> group the Hemochron times varied between 400 and 800 s during bypass. The patients receiving heparin alone showed a very significant lower Hemochron time ( $p < 0.01$  in early bypass; figure 6). Some of the placebo group of patients fell below 400 s towards the end of bypass and required an extra reinforcing dose of heparin with a correspondingly increased dose of protamine.

The thrombin times were slightly raised in the PGI<sub>2</sub> group from the time of administration of the heparin dose until 6 h after bypass, when they became the same as in the heparin group.

Platelet counts were consistently higher throughout bypass in the PGI<sub>2</sub> patients than in the placebo group ( $p < 0.01$ ). Figure 7 relates the platelet count and the platelet aggregation.

Platelet aggregation was reduced to 20% of initial values by PGI<sub>2</sub> throughout the period of its administration. In the prostacyclin group the platelet aggregation had recovered to 70% of the initial value 2 h after bypass. In the placebo group, platelet aggregability decreased gradually during bypass to approximately 30% of initial values and only partly recovered 2 h after bypass. By 24 h after bypass there was no significant difference.

TABLE 2. SUMMARY OF CLINICAL MEASUREMENTS  
(means  $\pm$  s.e.m.)

	PGI <sub>2</sub>	placebo
mean blood pressure after cannulation/mmHg		
systolic	81 $\pm$ 6	123 $\pm$ 9
diastolic	55 $\pm$ 4	88 $\pm$ 5
mean heart rate/min <sup>-1</sup>		
after cannulation	85 $\pm$ 5	93 $\pm$ 5
after protamine given	96 $\pm$ 6	90 $\pm$ 4
mean blood usage/ml		
during operation	596 $\pm$ 226	909 $\pm$ 260
after operation (18 h)	1073 $\pm$ 200	1495 $\pm$ 311
total across operation	1669 $\pm$ 303	2404 $\pm$ 485
blood loss/ml		
0-6 h	250 $\pm$ 29	499 $\pm$ 103
6-18 h	103 $\pm$ 8	273 $\pm$ 67
> 18 h	132 $\pm$ 18	138 $\pm$ 23
total	485 $\pm$ 48	910 $\pm$ 172
mean administration of other fluids/ml		
during operation	3095 $\pm$ 357	3384 $\pm$ 471
after operation (18 h)	841 $\pm$ 46	846 $\pm$ 44
total	3936 $\pm$ 346	4211 $\pm$ 454
mean urine output/ml		
during operation	1120 $\pm$ 108	1435 $\pm$ 229
after operation	1992 $\pm$ 337	1637 $\pm$ 115
total	3112 $\pm$ 426	3072 $\pm$ 221

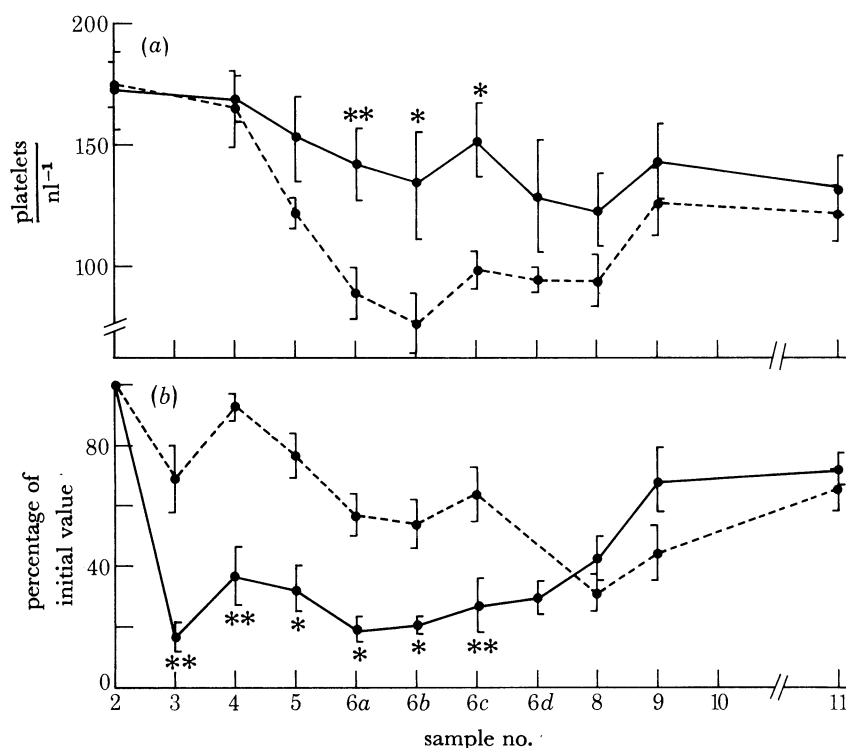


FIGURE 7. Variation of platelet count (a) and platelet aggregability (b) to ADP with (—) and without (---) PGI<sub>2</sub>. Note the protection of the platelet population in the presence of PGI<sub>2</sub>, and also that their aggregability is markedly diminished during bypass, recovering to *ca.* 70% 6 h later, whereas in the placebo group it is only 45% at this critical post-operative time. \*,  $p < 0.01$ ; \*\*,  $p < 0.05$ ; bars indicate standard error.

*Clinical measurements*

The clinical measurements are summarized in table 2. As expected, prostacyclin induced some hypotension, but this caused no difficulties in the clinical management of the patients. Only two patients required small doses of vasopressors. Our protocol allowed for any fall in blood pressure to be treated by the administration of additional fluid. In fact less fluid was required by the PGI<sub>2</sub> group. In spite of the lower blood pressures before bypass in the PGI<sub>2</sub> groups, the urine outputs were not significantly different in the two groups.

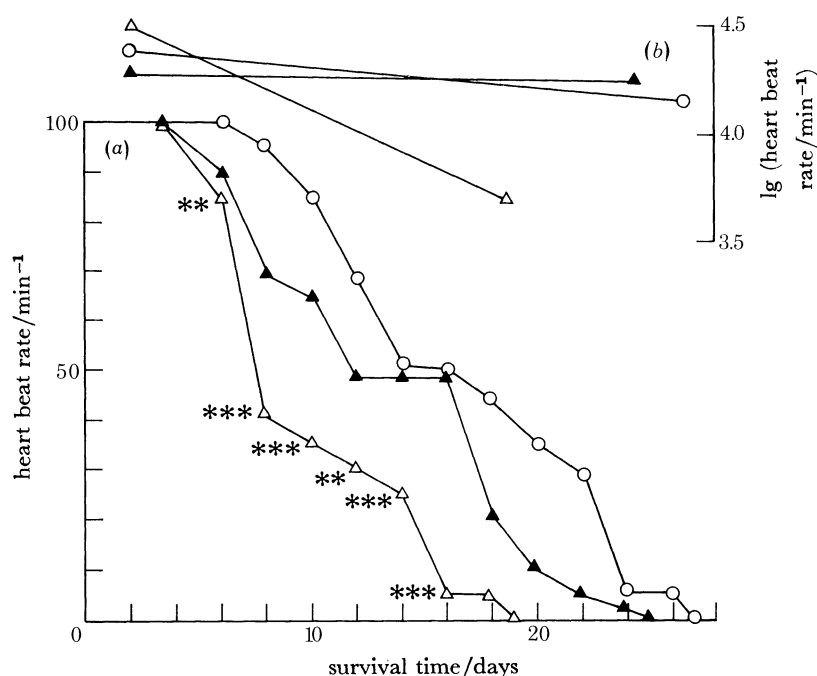


FIGURE 8. Reduction of heart rate and survival time (a) of cultured foetal mouse hearts; (b) linear regression analysis. The hearts exposed to plasma from the placebo group ( $\Delta$ ) show a significant depression of beating rate and survival time compared with that from a normal control ( $\circ$ ) and the PGI<sub>2</sub> group ( $\blacktriangle$ ). \*\*,  $p < 0.05$ ; \*\*\*,  $p < 0.001$ .

We were unable to detect significant differences in the cerebral function and neurological status between the two groups of patients. No patient in either group showed any permanent detectable psychopathological change. All became more skilful with the repeated tests, showing an apparent improvement by 6 days after operation. Importantly, over half of the placebo group complained of inability to focus their eyes and to read newspaper print for 48 h. None of the PGI<sub>2</sub> group suffered from this disability.

The foetal heart culture test showed a significant depression of the foetal hearts exposed to plasma taken from blood samples no. 7 at the end of bypass in the placebo group, with reduction of the beating rate and survival time ( $p < 0.001$ ) as shown in figure 8. Median survival times ( $l.t._{50}$ ) showed no significant difference between the foetal hearts exposed to plasma proteins from the PGI<sub>2</sub> group and plasma obtained from a normal volunteer, whereas there were marked differences with the placebo sample. The creatinine clearance test showed no significant difference between the two groups.

## DISCUSSION

The three known effects of prostacyclin were confirmed with this study: stabilization of the platelet in the extracorporeal circulation, the heparin sparing effect and the vasodilator effect. In addition to these effects of PGI<sub>2</sub>, our studies suggest that PGI<sub>2</sub> reduces the capillary damage leading to increased capillary permeability that takes place during cardiopulmonary bypass. The mechanism of this may be related to the reduction of platelet granule release. This mechanism is suggested by the foetal heart studies.

*Foetal heart test*

The foetal hearts showed significantly better survival times and beating performance when exposed to plasma from patients who had received PGI<sub>2</sub> compared with those who had received placebo. This is probably due to a reduction in the amount of platelet granular substances liberated when prostacyclin is administered. In our previous studies without the use of PGI<sub>2</sub> (Longmore *et al.* 1980) we have always shown depression of beating rate and survival time in foetal hearts when they are exposed to plasma taken at the end of bypass. The corresponding plasma from the PGI<sub>2</sub> patients was comparable with the control blood drawn from the healthy volunteer. It has been argued that the cause of the depression of the foetal hearts that follows a normal bypass may be associated with mechanical damage to proteins or may be due to protein damage following granule release from platelets. The beneficial effects of PGI<sub>2</sub> suggests the latter mechanism, as does the possible change in capillary permeability.

The  $\beta$ -thromboglobulin results do not help to resolve this problem. Plasma  $\beta$ -thromboglobulin concentration was increased in samples from both groups during and after surgery. This suggests a high level of platelet activation, but massive *in vitro* liberation of  $\beta$ TG may take place in the long cannulae through which blood samples were withdrawn.

*Platelet aggregation and reduction of blood loss*

The PGI<sub>2</sub> effect on platelet aggregation in the human clinical trial was less profound than in the experiments on dogs. Nevertheless, there was virtually complete preservation of the platelet population throughout cardiopulmonary bypass. Importantly, the platelet function returned almost to normal after bypass with a concomitant halving of blood loss. No patient in the PGI<sub>2</sub> series bled sufficiently to cause anxiety post-operatively. The total blood loss was so small that administration of blood after bypass would appear to be superfluous when PGI<sub>2</sub> is used.

*Heparin sparing effect*

The heparin sparing effect previously described in experimental renal dialysis by Woods *et al.* (1978) was confirmed. The effect of this was to reduce the amount of additional heparin required towards the end of bypass to maintain an acceptable Hemochron level. The amount of protamine required in the PGI<sub>2</sub> group was correspondingly less.

*The vasodilator effect of PGI<sub>2</sub>*

The vasodilator effect of the pre-perfusion dose of 10 ng kg<sup>-1</sup> min<sup>-1</sup> was variable. It was troublesome enough to indicate the need for small doses of vasopressor agents in only two patients. In all other cases in this series, the anaesthetist was unable to determine whether prostacyclin was present although he was looking for hypotension and facial flushing. We

expected that the hypotensive effects of PGI<sub>2</sub> might require the administration of more fluids during bypass to maintain the blood pressure; even when the dose of PGI<sub>2</sub> was doubled to 20 ng kg<sup>-1</sup> min<sup>-1</sup> at the beginning of bypass this was not so. There was no significant difference between the flow rates in the two groups, although it is our practice to increase the flow rather than administer vasopressors when the perfusion pressures are low.

We also considered the theoretical possibility that any hypotension before the bypass started could cause a reduction in renal perfusion and urine flow. If this was followed by an increase in urine flow towards the end of bypass, it might be difficult to administer sufficient potassium in the depleted patient. We did not experience this problem. In two patients in the PGI<sub>2</sub> group, the urine flow was reduced before bypass, but within the first few minutes of bypass, urine started to flow. No difficulties in the management of the serum potassium was experienced in these patients.

In this series, Bos 10 bubble oxygenators were used because we were not certain what the effects of PGI<sub>2</sub> would be on the deposition of the layer of blood products on the surface of 'Celgard' microporous membranes upon which oxygenation depends in some membrane oxygenators. In a subsequent pilot study of membrane oxygenators, using dogs, we find by scanning electron micrography that in spite of the presence of prostacyclin, this essential layer is deposited although it is thinner. There would appear to be no contra-indication to the use of PGI<sub>2</sub> with membrane oxygenators. We are investigating this further.

We were disappointed that the reduction in the amount of brain damage that sometimes occurs in cardiopulmonary bypass owing to platelet aggregates and intravascular clotting was not obvious in this trial. We used the memory recall tests devised by Bethune (1980). In all our patients, 3 days after operation there was a demonstrable impairment of cerebral function, but by 6 days after operation no patient showed any cerebral dysfunction according to the test. Bethune now feels that the test is not sufficiently sensitive for this purpose. The International Group (Katz *et al.* 1978) studying cerebral damage after open-heart surgery is proposing to use a combination of memory recall, conceptual logic analogue testing and word rotation tests; this is more sensitive. We obtained some difficulty in obtaining the patients' attention in the busy pre-operative period and feel that a long and complex test is undesirable and might distress the patient at that time. Over half of the placebo patients complained of visual disturbances that were not experienced by the PGI<sub>2</sub> group. Further investigation is required to find whether PGI<sub>2</sub> will eliminate all multi-organ and cerebral complications.

#### CONCLUSION

In spite of the theoretical possible disadvantages of the vasodilator effects of PGI<sub>2</sub> referred to by Salzman (this symposium), both animal studies and a carefully conducted double-blind human clinical trial show them to be irrelevant and far outweighed by the real measured advantages of its use.

Exploitation of the beneficial effects of PGI<sub>2</sub> in extracorporeal circulation probably presents the most important potential advance in open-heart surgery in the 23 years since it first became routine.

There are, however, two coincidental advances, which probably make the discovery of PGI<sub>2</sub> even more important. These are in the unrelated field of computer-enhanced non-invasive instruments for the early diagnosis of cardiovascular disease (Longmore *et al.* 1976) and the

discovery of growth-limiting substances of the arterial wall smooth muscle cells (Florentin *et al.* 1973; Nam *et al.* 1974; Thomas *et al.* 1976). We may soon enter a new era in the management of cardiovascular disease, with early diagnosis of atherosclerosis, inhibition of untoward platelet activity blocking vessels and stimulating smooth muscle proliferation (Stemerman 1979), and control of hyperplastic smooth muscle. PGI<sub>2</sub> is an important first member of a family of substances that may well enable medicine to influence the progress of the disease process which causes over half of all deaths and morbidity in the Western World.

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#### REFERENCES (Longmore)

- Åberg, T. 1974 Effect of open heart surgery in intellectual function. *Scand. J. thorac. cardiovasc. Surg., Suppl.* **15**, 1–63.
- Bass, R. M. & Longmore, D. B. 1969 Cerebral damage during openheart surgery. *Nature, Lond.* **222**, 30–33.
- Bethune, D. W. 1980 Psychometric testing in the evaluation of post-operative cardiac patient. In *Towards safer cardiac surgery* (ed. D. B. Longmore), pp. 613–617. Medical Technical Press.
- Born, G. V. R. 1962 Aggregation of blood platelets by adenosine diphosphate and its reversal. *Nature, Lond.* **194**, 927–929.
- Ellison, N., Edmunds, L. H., Jr & Colman, R. W. 1978 Platelet aggregation following heparin and protamine administration. *Anesthesiology* **48**, 65–68.
- Florentin, R. A., Nam, S. C., Janakidevi, K., Lee, K. T., Reiner, J. M. & Thomas, W. A. 1973 Population dynamics of arterial smoothmuscle cells. II. *In vivo* inhibition of entry into mitosis of swine arterial smooth-muscle cells by aortic tissue extracts. *Arch. Path.* **95**, 317–320.
- Hope, W., Martin, T. J., Chesterman, C. N. & Morgan, F. J. 1979 Human  $\beta$ -thromboglobulin inhibits PGI<sub>2</sub> production and binds to a specific site in bovine aortic endothelial cells. *Nature, Lond.* **282**, 210–212.
- Hughes, D. M. & Longmore, D. B. 1972 Relationship between the stage of development of foetal hearts and their survival in organ culture. *Nature Lond.* **235**, 334–336.
- Katz, J., Longmore, D. B. & Speidel, H. 1980 *Secretariat, International Study Group on Cerebral and Psychopathological Dysfunction following Cardiac Surgery* Milwaukee County Mental Health Complex, 9191 Watertown Plank Road, Milwaukee, Wisconsin 53226, U.S.A.
- Larkin, J. D., Blocker, T. J., Strong, D. M. & Yocum, M. W. 1978 Anaphylaxis to protamine sulphate mediated by complement-dependent IgG antibody. *J. Allergy clin. Immunol.* **61**, 102–107.
- Longmore, D. B. & Smith, M. A. 1980 Toxins in open heart surgery. In *Towards safer cardiac surgery* (ed. D. B. Longmore), pp. 447–473. Medical Technical Press.
- Longmore, D. B., Bennett, G., Gueirrara, D., Smith, M., Bunting, S., Moncada, S., Reed, P., Read, N. G. & Vane, J. R. 1979 Prostacyclin: a solution to some problems of extracorporeal circulation. *Lancet* **i**, 1002–1005.
- Longmore, D. B., Bennett, G., Hoyle, P. M., Smith, M. A., Gregory, A., Osivand, T. & Jones, W. A. 1981 Prostacyclin administration during cardiopulmonary bypass in man. *Lancet* **i**, 800–804.
- Longmore, D. B., Turk, J. L. & Lockey, E. 1978 *Scientific Manifesto of H.E.A.R.T.*, 47 Wimpole Street, London, W.1, U.K.
- Ludlam, C. A., Moore, S., Bolton, A. E., Pepper, D. S. & Cash, J. D. 1975 The release of a human platelet specific protein measured by a radioimmunoassay. *Thromb. Res.* **6**, 543–548.

- Moncada, S. & Vane, J. R. 1978 Unstable metabolites of arachidonic acid and their role in haemostasis and thrombosis. *Br. med. Bull.* **34**, 129–135.
- Nam, S. C., Florentin, R. A., Janakidevi, K., Lee, K. T., Reiner, J. M. & Thomas, W. A. 1974 Population dynamics of arterial smooth-muscle cells. III. Inhibition by aortic tissue extracts of proliferative response to intimal injury in hypercholesterolemic swine. *Expl molec. Pathol.* **21**, 259–267.
- Stemerman, M. B. 1979 Hemostasis, thrombosis and atherogenesis. *Atherosclerosis Rev.* **6**, 105–146.
- Thomas, W. A., Janakidevi, K., Florentin, R. A., Lee, K. T. & Reiner, J. M. 1976 Search for arterial smooth-muscle cell chalone(s). Chapter 20 In *Chalones* (ed. J. H. Houck), ch. 20. New York: North-Holland.
- Velders, A. J., van den Dungen, J. J. A. M., Westerhof, N. J. W. & Wildevuur, C. R. H. 1980 Platelet damage by protamine administration: protection by reducing protamine or by prostacyclin (PGI<sub>2</sub>) treatment. *Eur. surg. Res.* **12** (suppl. 1), 50–51.
- Wildenthal, K. 1971 *a* Factors promoting the survival and beating of intact foetal mouse hearts in organ culture. *J. molec. Cell Cardiol.* **1**, 101–104.
- Wildenthal, K. 1971 *b* Long-term maintenance of spontaneously beating mouse hearts in organ culture. *J. appl. Physiol.* **30**, 153–157.
- Wolf, P. 1967 The nature and significance of platelet products in human plasma. *Br. J. Haemat.* **13**, 269–288.
- Woods, H. F., Ash, G., Weston, M. J., Bunting, S., Moncada, S. & Vane, J. R. 1978 Prostacyclin can replace heparin in haemodialysis in dogs. *Lancet* ii, 1075–1077.



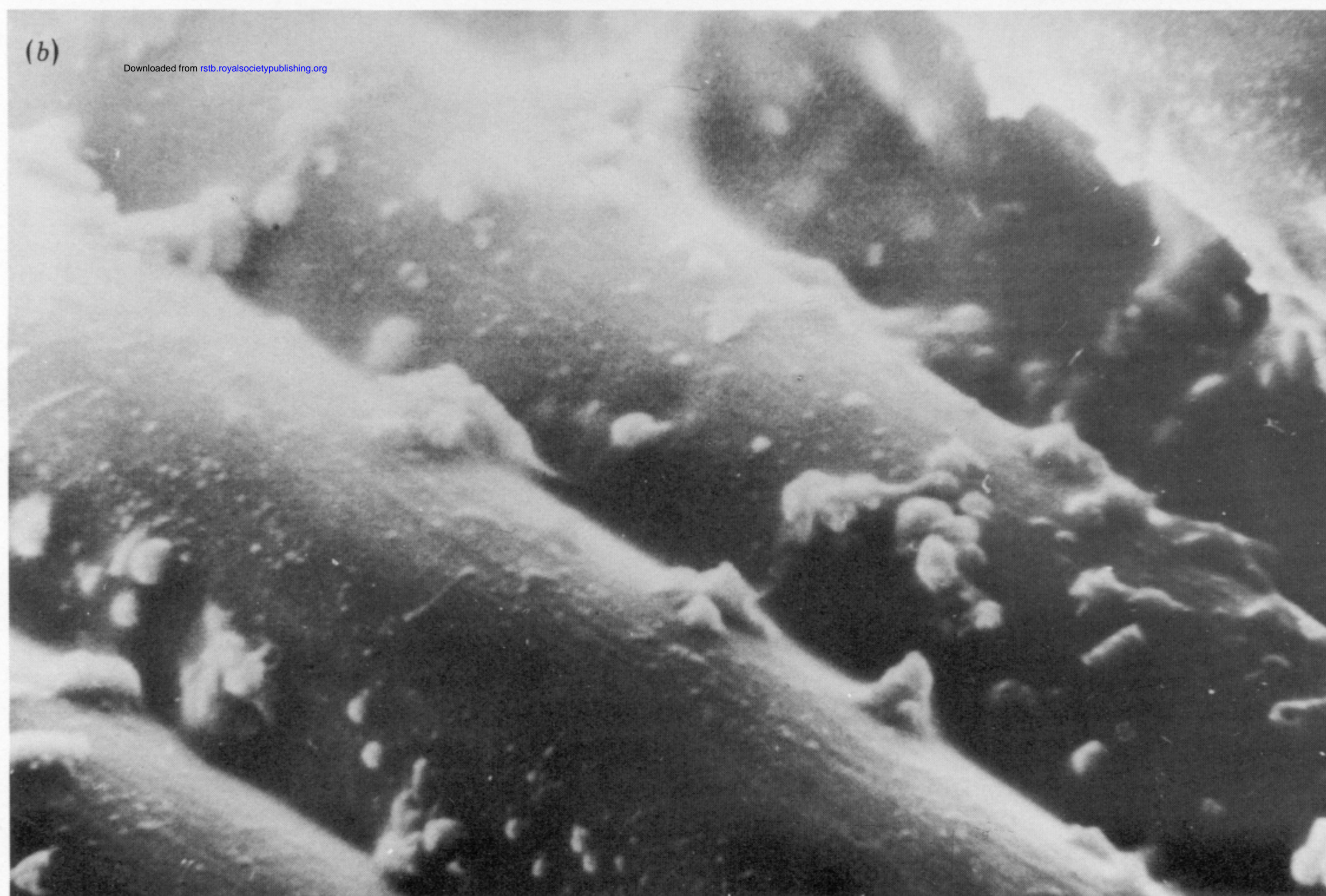
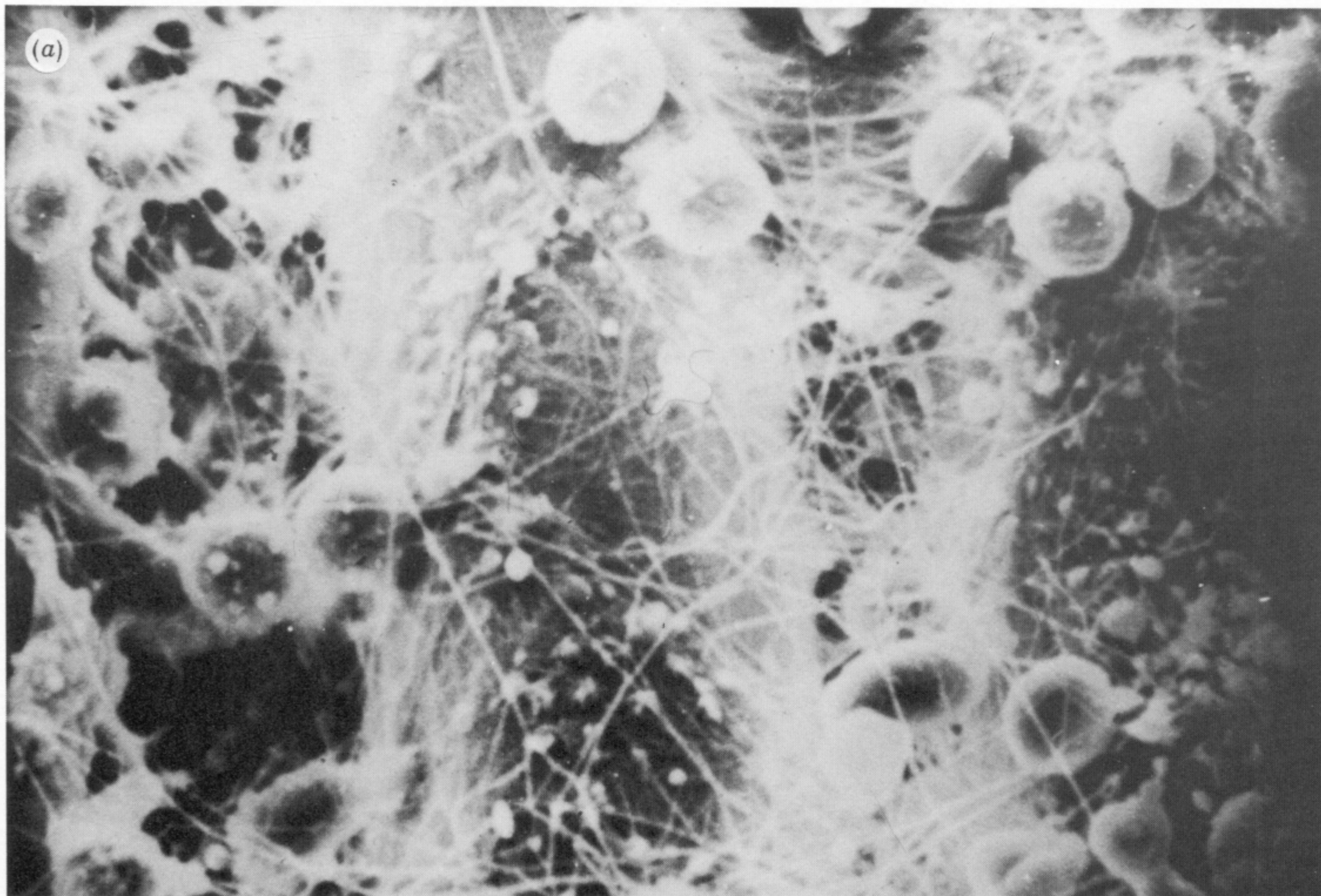


FIGURE 4. (a) Scanning electron microscopy showed a build-up of platelet aggregates, fibrin strands and red and white cells on the filter from the bypass with heparin alone. (b) Scanning electron microscopy showed few platelet aggregates and no fibrin strands when  $\text{PGI}_2$  was used in combination with heparin.