In other words, if 10 ng of PGE_1 per milliliter of plasma is used in 250 ml of PRP, only 0.25 μg, a negligible amount, will be transfused into the recipient, where it is known to be rapidly metabolized (*14*).

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Prostaglandin E_1 in Preparation and Storage of Platelet Concentrates

Abstract. *The addition of prostaglandin E_1 to blood collection bags improved platelet harvesting; platelets were easily suspended immediately after centrifugation. The treatment with prostaglandin E_1 did not affect the survival time of these platelets after infusion into a recipient.*

At the recommendation of Shio and Ramwell (*1*), we studied the effects of prostaglandin E_1 (PGE_1) in the preparation of and short-term preservation of platelets.

One milligram of PGE_1 (*2*) was dissolved in 0.2 ml of ethanol and diluted with isotonic saline to 20 ml (final concentration, 50 ng/μl). The solution was sterilized by filtration through a 0.2-μm filter (Millipore) and stored in a sterile container at 4°C until use. Its potency was checked periodically by testing its ability to inhibit adenosine diphosphate (ADP)–induced aggregation of fresh normal platelets. Platelet morphology was studied by phase microscopy of wet platelet suspensions and by electron microscopy (*3*) of platelets fixed with glutaraldehyde and osmium tetroxide. The amount of platelet aggregation was determined with an aggregometer (Chrono-Log) and platelet viability in vivo was determined by survival studies with ^{51}Cr (*4*). Blood was collected in Fenwal triple packs (FP-297). Platelet-rich plasma (PRP) was prepared by centrifuging whole blood at 1500*g* for 6 minutes. Plasma was then transferred to a second bag and centrifuged at 4000*g* for 10 minutes to compact the platelets; all but 30 ml of plasma was expressed into the third pack. The platelet concentrate was suspended and stored on a rotator which maintained a state of gentle agitation (*5*).

In 24 studies in which PGE_1 (4 to 200 μg per bag) was added to either whole blood or PRP, clumping of platelets was totally prevented in concentrates that were prepared either at room temperature or at 4°C, and either with acid, citrate, and dextrose (ACD) or with citrate, phosphate, and dextrose (CPD) anticoagulants (Fig. 1, a and b). Concentrates thus prepared could be suspended with only a minimum of manipulation immediately after rapid centrifugation. The smallest quantity of PGE_1 used (12 ng of PGE_1 per milliliter of plasma) was about one-fourth the amount needed to produce 50 percent inhibition of aggregation (induced by $2 \times 10^{-6} M$ ADP) of fresh platelets.

Platelet viability and function were

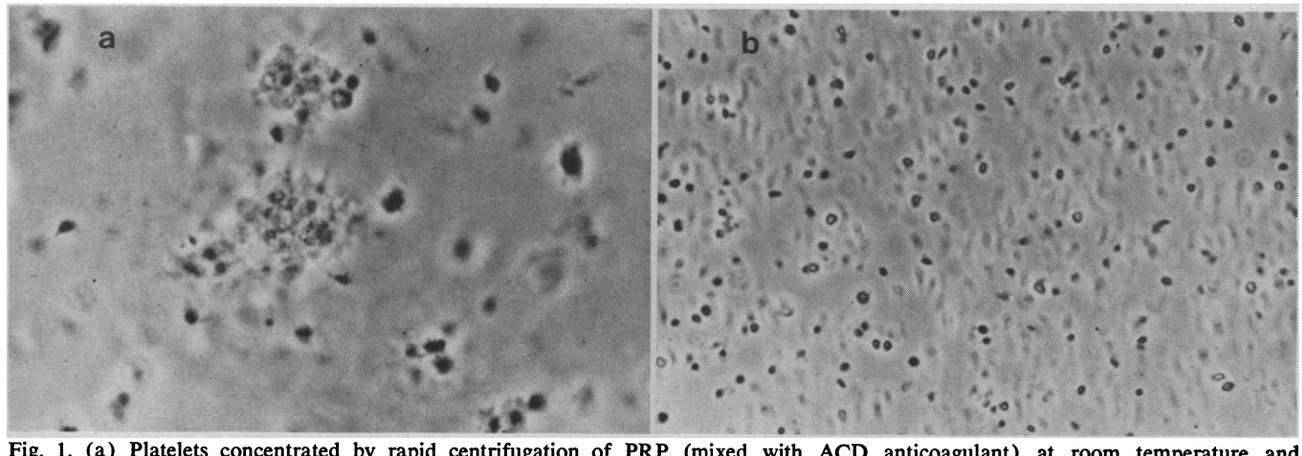

Fig. 1. (a) Platelets concentrated by rapid centrifugation of PRP (mixed with ACD anticoagulant) at room temperature and then immediately suspended. Many clumps of platelets were present which did not disperse completely even after 2 days of gentle agitation at room temperature. About 50 percent of the total platelets were contained in the aggregates. At 5°C, or when CPD anticoagulant was used, platelet clumping was even more pronounced (× 980). (b) Platelets prepared as in (a) but with the addition of 8 μg of PGE_1 to PRP before rapid centrifugation (final PGE_1 concentration, 24 ng/ml). A smooth suspension of platelets was obtained within 15 to 30 seconds (× 400).

studied with platelets from concentrates prepared with PGE_1. They were diluted, to a final concentration of 300,000/mm^3 (about 1:10), in ABO-compatible, citrated, platelet-poor plasma. Aggregation in response to ADP ($2 \times 10^{-6}M$), collagen, and epinephrine ($10^{-5}M$) was normal. The survival times (measured with ^{51}Cr) of platelets that were concentrated and then immediately suspended in PGE_1 were also normal (Fig. 2).

Platelet clumping was inhibited by PGE_1 (12 to 750 ng/ml) in whole blood that was stored at 4°C; 50 to 100 percent of the platelets could be recovered even after the blood was chilled for 24 hours. Suspension of concentrated platelets was almost immediate in samples having the greatest concentrations of PGE_1, and with lesser concentrations, up to 30 minutes was required. Platelets obtained in this way were spherical in shape, but otherwise had normal structure. No additional change in shape occurred after incubation at 37°C for 1 hour. Aggregation in response to ADP, collagen, and epinephrine was the same as that of fresh platelets. Recovery (percent of total platelets remaining in the circulation 1 hour after injection) of such platelets labeled with ^{51}Cr was normal on transfusion, but their survival time was short (half-life, 1.5 days) and comparable to that of platelet concentrates stored for the same period of time at 4°C.

To determine whether PGE_1 might aid in short-term preservation of platelets, we performed 36 studies with platelet concentrates containing 50 to 700 ng of PGE_1 per milliliter. After the addition of PGE_1, the concentrates were stored for 24 to 120 hours, either at room temperature or at 4°C. In the presence of PGE_1, the change of shape of the platelet from a disc to a sphere was delayed, but it was not possible to demonstrate that platelet ultrastructure and function were better preserved with PGE_1 than they were in the absence of the drug. These were also the results of four studies in which 50 μg of PGE_1 was added daily to the concentrates previously mixed with the drug to compensate for inactivation of PGE_1 during storage. Survival studies with ^{51}Cr were performed only on platelets stored at 4°C with PGE_1 (250 ng/ml). Survival times of these platelets were uniformly short and were comparable to those of platelets stored without PGE_1 at 4°C for the same period of time.

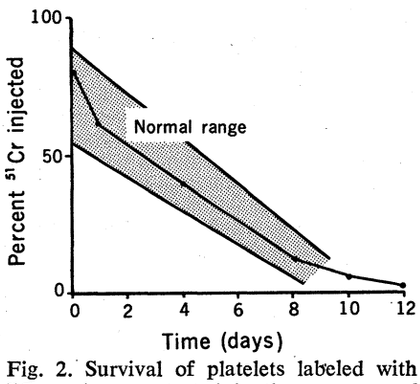

Fig. 2. Survival of platelets labeled with ^{51}Cr and concentrated in the presence of PGE_1 (24 ng/ml).

Platelet concentrates prepared with 250 ng of PGE_1 per milliliter to facilitate suspension do not tolerate freezing (*6*) any better than untreated platelets do.

Concentrations of PGE_1 as low as 24 μg/ml permit platelet pellets to be suspended almost immediately after rapid centrifugation with no adverse effect on their function or survival. Platelets similarly treated, but with no added PGE_1, clump irreversibly. Thus, addition of small amounts of PGE_1 at the time whole blood is collected reduces by 1 to 2 hours the time required for preparation of platelet concentrates. The addition of PGE_1 may also improve the quality of the final product by preventing even microscopic clumping of platelets.

Addition of as little as 4 μg of PGE_1 to a unit of whole blood at the time of collection permits 50 to 100 percent of the platelets initially present to be recovered in platelet concentrates, even after 24 hours of storage at 4°C. Although the survival time of such platelets is short, they appear to function normally in vitro. Clinical studies indicate that they are effective in stopping hemorrhage in thrombocytopenic patients. Although a concentration of PGE_1 of 12 ng/ml is insufficient to inhibit ADP-induced platelet aggregation, it does prevent platelet clumping in stored whole blood.

One of the factors limiting the availability of platelets for therapeutic purposes is that much of the blood used for transfusion is collected at mobile sites and must, by regulations of the National Institutes of Health and the American Association of Blood Banks, be "chilled immediately" upon collection. By the time this blood has been transported to the processing center, recovery of platelets in the form of concentrates is virtually impossible. Use of PGE_1 may permit recovery of platelets suitable for transfusion from such blood. The maximum delay permissible before platelets are extracted from chilled blood to which PGE_1 has been added, without shortening their survival time in vivo, remains to be determined.

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Prostaglandins in the Preparation of Blood Components

Abstract. *Prostaglandin E_1 significantly improved the separation of blood components in blood bags. The recovery in vivo and the life-span values of the platelets were not altered. The hemostatic effectiveness of platelets treated with prostaglandin was shown to be normal in man.*

At the suggestion of Shio and Ramwell (*1*), we carried out experiments in which prostaglandin E_1 (PGE_1) was added to whole blood that was collected in the anticoagulant citrate-phosphate-dextrose (CPD) and to the platelet-rich plasma (PRP) prepared from this blood. We studied the effects of this procedure on (i) recovery in vitro of platelets from whole blood, (ii) the survival in vivo of both liquid-stored and previously frozen platelets, (iii) the survival in vivo of liquid-stored red cells, and (iv) physical, structural, and metabolic parameters of human red cells stored with PGE_1 and PGE_2 (*2*).