A MODEL OF DISSEMINATED INTRAVASCULAR BLOOD CLOTTING

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Vascular-platelet and plasma hemostasis was studied in dogs after blood loss (40-45 ml/kg body weight) followed by hypervolemic (60-65 ml/kg body weight) transfusion of homologous (from three donors) plasma enriched with platelets and leukocytes. After a brief phase of hypercoagulation, hypocoagulation was discovered in all the experimental dogs, accompanied by a reduction in the platelet count and in the intensity of platelet aggregation, lengthening of the bleeding time, a decrease in the resistance of the capillary wall, a decrease in the plasma fibrinogen concentration and activity of factor XIII, and an increase in the fibrinolytic activity of the blood. The results are evidence of the development of an acute syndrome of disseminated intravascular blood clotting.

KEY WORDS: plasma transfusion; disseminated intravascular blood clotting.

Disseminated intravascular blood clotting (DIBC), or the thrombohemorrhagic syndrome is nowadays recognized to be a universal general pathological process, for it is observed in various types of clinical pathology [1, 3, 4, 9, 10]. The widespread occurrence of the DIBC syndrome in clinical medicine, the different varieties and forms of its manifestation, but at the same time, the many unsolved problems in its pathogenesis have naturally aroused the interest of research workers. To study the mechanisms of development of DIBC many experimental models have been created [1] that adequately reflect certain forms of special pathology. A DIBC syndrome can be found after transfusion of massive doses of stored blood or of packed platelets and leukocytes. Meanwhile, under experimental conditions, no adequate model yet exists.

An investigation was accordingly conducted with the object of creating a new experimental model of DIBC.

EXPERIMENTAL METHOD

Experiments were carried out on five male mongrel dogs subjected, under pentobarbital anesthesia, to acute blood loss (40-45 ml/kg body weight) followed by transfusion of fresh plasma containing a suspension of platelets and leukocytes, made up on the day of the experiment from three donor dogs. Blood of the recipient and donor dogs was matched for group erythrocyte antigens. The volume of plasma transfused amounted to 150% of the volume of blood removed; the transfusion began with a continuous jet (about 50% of the volume to be transfused), and later by rapid drip. The duration of transfusion was 20-25 min. Together with plasma, in the course of transfusion the experimental dog also received 120-140 million platelets and about 2 million leukocytes/kg body weight.

Blood samples were taken from the femoral vein before bleeding (initial state), 5 min after the beginning of transfusion, at its end, and 1, 2, and 4 h after transfusion.

To characterize the coagulation properties of the blood the following indices were studied: of plasma hemostasis – the thromboelastogram (TEG), plasma fibrinogen concentration [7], fibrinolytic activity of the blood [6], fibrinogen degradation products [8], and plasma factor XIII [2]; of vascular-platelet hemostasis – the platelet count (using a counting chamber), the adhesive activity of the platelets [5], aggregation of the platelets under the influence of ADP [5], the bleeding time, and the resistance of the capillary wall (Nesterov's test).

EXPERIMENTAL RESULTS

All the experimental dogs developed the picture of a severe post-transfusion complication: dyspnea, convulsions, hyperemia of the skin, and petechiae on the skin and mucous membranes. Of the five dogs one

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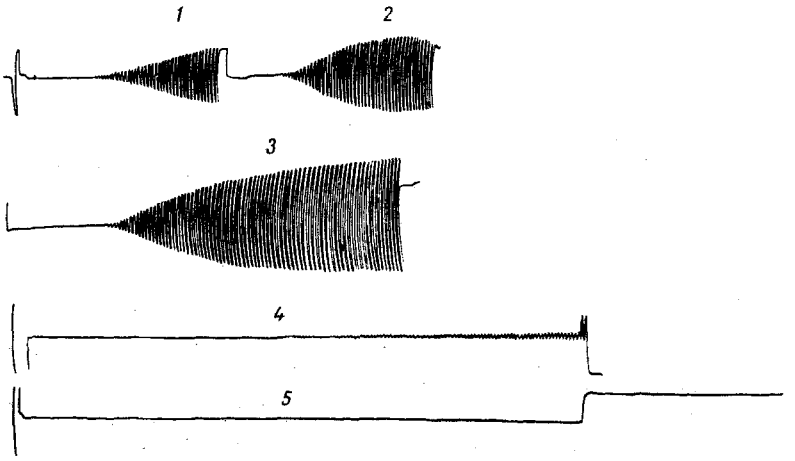

Fig. 1. Dynamics of changes in clotting power of blood in dog No. 3 after transfusion of plasma enriched with platelets and leukocytes (data of the TEG). P) Thromboelastographic constant of thromboplastin; K_1) thromboelastographic constant of thrombin; P + K) nonspecific coagulation constant; T_M) constant of specific blood clotting. 1) TEG in initial state (before transfusion): P = 35 mm, K_1 = 40 mm, P + K = 75 mm, T_M = 10 mm; 2) TEG 5 min after beginning of transfusion; hypercoagulation with signs of commencing hypocoagulation: P = 25 mm, K_1 = 20 mm, P + K = 45 mm, T_M = 25 mm; 3) TEG at end of transfusion – well-marked hypocoagulation: P = 45 mm, K_1 = 30 mm, P + K = 75 mm, T_M = 100 mm; 4-5) TEG 1-2 h after plasma transfusion: complete inability of blood to clot.

survived, one was killed 2 h later in a serious condition, and three died after 4, 6, and 24 h – one of them from massive bleeding from a small wound in the tongue caused during intubation, and one also from bleeding from wound sutures. The clinical manifestations of the post-transfusion complication were the same in all the animals, and only the severity of the reaction varied.

Changes in the clotting system of the blood of one dog are illustrated in Fig. 1. The more rapid formation of thromboplastin and thrombin and conversion of fibrinogen into fibrin (TEG) was found 5 min after the beginning of transfusion (Fig. 1, 2). Compared with the initial state (Fig. 1, 1), the TEG2 showed evidence of hypercoagulation, although there were some signs of impending hypocoagulation (Fig. 1, 3). Well-marked hypocoagulation was present 1 h after transfusion (Fig. 1, 4), and changed into complete inability of the blood to clot during the subsequent period (Fig. 1, 5). Similar changes were present in all the dogs.

The fibrinogen concentration (Fig. 2, 1) fell throughout the period of transfusion to 30-40% of its initial level, and remained at about the same value for the next 4 h. The fibrinolytic activity of the blood (Fig. 2, 2) increased actually during transfusion, and 1-2 h thereafter it was 50-60% higher than initially. Fibrinogen degradation products appeared 1 h after the end of transfusion (Fig. 2, 3), and after 4 h their level in the peripheral blood was doubled. Plasma factor XIII activity fell during transfusion in four experiments, and the decrease was greatest (by 50%) 2 h after transfusion.

The platelet count (Fig. 3, 1) was reduced by half at the very beginning of transfusion (despite the excess of platelets in the transfused plasma). The platelet count 1-2 h after the end of transfusion was 40-50% of its initial level, and it remained at the same level (50%) later. The adhesive function of the platelets (Fig. 3, 2) increased during transfusion, but fell to its initial level 1 h after the end of transfusion, below its initial level after 2 h, and rose again after 4 h. Aggregation of the platelets (Fig. 3, 3) was reduced by 80% 5 min after the beginning of transfusion, it continued to decrease later, and could not be determined 4 h after the end of transfusion. The bleeding time increased during transfusion and the first hour thereafter (Fig. 3, 4). Changes in this index correlated with changes in the adhesive function of the platelets. Nesterov's test was positive in three of five experiments 2 and 4 h after transfusion.

At autopsy on one of the dogs which died from bleeding, massive hemorrhages were found into the heart muscle and the tissues of the lungs, liver, spleen, and kidneys. Large juxtamural thrombi and multiple blood

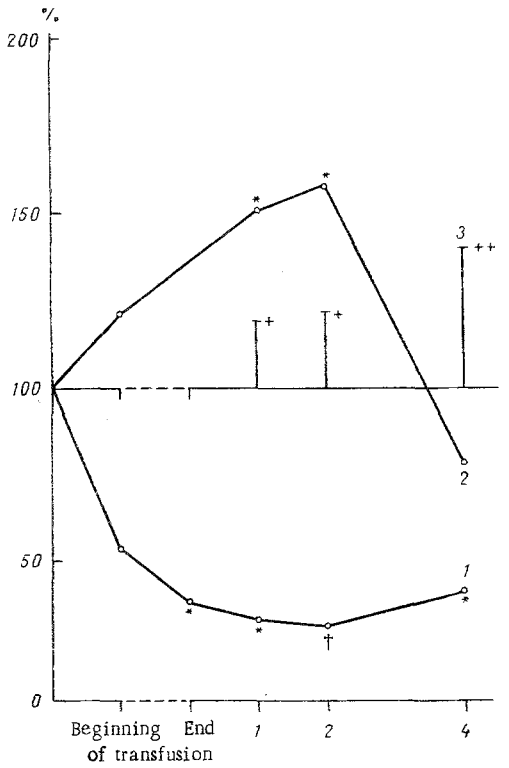

Fig. 2

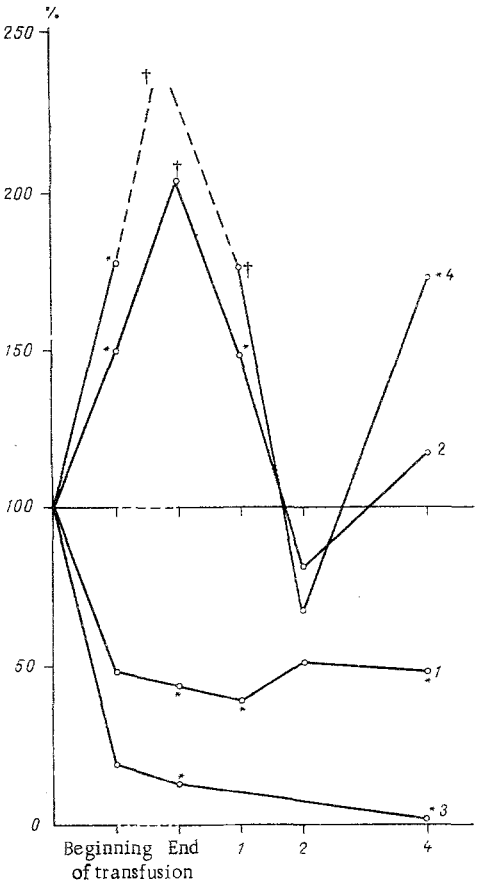

Fig. 3

Fig. 2. Changes in fibrinogen concentration (1), fibrinolytic activity (2), and fibrinogen degradation products (3) after transfusion of homologous plasma. Abscissa, time after transfusion of plasma (in h); ordinate, changes in indices studied (in % of initial level, taken conventionally as 100%). Asterisks and dagger denote significance of differences between initial values and values determined at corresponding times: *) $P < 0.05$; †) $P < 0.01$.

Fig. 3. Changes in platelet count (1), adhesive (2) and aggregation (3) functions of platelets, and bleeding time (4) after transfusion of homologous plasma. Remainder of legend as in Fig. 2.

clots were found in the large blood vessels, despite the total loss of the clotting power of the blood. Multiple hemorrhages of varied severity were found in the other dogs also at autopsy.

It can be concluded from these results that the transfusion of massive doses of donors' plasma enriched with platelets and leukocytes, after acute blood loss, leads to the development of a well-marked DIBC syndrome, accompanied by disturbance of vascular-platelet hemostasis (a reduction in the number and aggregation of the platelets, lengthening of the bleeding time, a decrease in the resistance of the capillary walls). Mobilization of platelets within microclots over extensive areas (of the lung and liver tissue, etc.) leads in turn to a reduction in the concentration of coagulation factors in the circulating blood.

Besides the disseminated formation of fibrin, the fibrinolytic activity of the blood also was increased, and as a result the residual fibrinogen and other procoagulants were broken down by proteolysis. All these factors account for the inability of the blood to clot.

The phase of hypercoagulation in most experiments was almost undetectable, and hypocoagulation appeared after a very short time. Possibly by varying the rate of plasma transfusion or the number of transfused cells states corresponding to the different phases of DIBC (hyper- and hypocoagulation) could be created. According to the results of these experiments a DIBC syndrome can arise following the transfusion of fresh as well as of stored blood. Analogous situations may perhaps also arise in transfusion practice when massive doses of blood or packed leukocytes and platelets are transfused (Table 1). The suggested model may prove

TABLE 1. Models of Experimental Intravascular Blood Clotting and Its Clinical Equivalents (table taken from Baluda [1] with the addition of the suggested model)

Model	Clinical equivalent
1. Foreign surface	Thrombophlebitis. Myocardial infarction
2. Thermal injury	Heat stroke. Burns
3. Stasis	Shock. Hemangiomas. Aneurysms. Cardiopulmonary failure
4. Injection of virus	Hemorrhagic fever
5. Injection of bacterial endotoxins	Septicemia
6. Injection of amniotic fluid	Embolism from amniotic fluid
7. Antigen–antibody reaction	Schwartzmann phenomenon. Immune diseases. Thrombocytopenic purpura
8. Injection of hemolyzed blood	Transfusion of incompatible blood
9. Injection of thrombin or thromboplastin	Separation of the placenta. Intrauterine death of the fetus. Abortion. Trauma. Operations with considerable tissue injury. Malignant neoplasms. Snakes bites. Shock (traumatic, infectious)
10. Transfusion of plasma enriched with platelets and leukocytes	Massive transfusions of fresh donors' blood. Transfusion of packed platelets. Transfusion of packed leukocytes

useful for the study of the pathogenesis of the DIBC arising in transfusion practice, and of the mechanisms of deposition and distribution of the donor's platelets in the recipient and their participation in thrombus-formation, etc. The model may also be suitable for the study of the action of various pharmacological agents, antiaggregants, blood and plasma substitutes, and transfusion media with specific action.

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