NEW DATA ON THE SITE OF HEMOPOIETIN FORMATION

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Although there have been numerous researches undertaken with the aim of elucidating the site of hemopoietin formation, so far they have yielded no definite results. Appropriate literature references have been given in a previous account [3].

It is well known that hemopoietins can be found in the blood under varied conditions of hypoxia including those following hemorrhagic anemia. We have shown [2] that immediately after a single, massive bleeding of dogs, the hemopoietin content of blood taken from the femoral vein is sharply reduced; 3-5 h later hemopoietin activity begins to increase and it is maintained at a high level over the 20-24 h following blood-letting, except for some variation as a result of the individual peculiarities of the subject.

In order to determine which of the various possible organs liberate hemopoietins into the blood, in this present work we have studied the hemopoietic activity of blood serum derived from various organs of dogs, following the stimulatory effect of a single massive blood-letting.

EXPERIMENTAL METHODS

Experiments were carried out on 15 male dogs weighing 10-33 kg. The femoral artery and vein of the experimental animals were first separated, after which their abdominal cavities were opened up and blood samples taken under aseptic conditions from the veins of the kidney, spleen, liver, stomach, and small intestine, and from the femoral arteries and veins, in order to study the hemopoietin content before hemorrhage. After this we allowed a volume of blood equivalent to 25-30 mg per kg body weight to flow from the femoral artery. One to 2 h later we took blood samples from the 7 vessels enumerated above and repeated this sampling 5 times. In this way 42 blood samples were taken from each dog. The hemopoietin activity of the blood serum was determined by a hemoculture method [1] using a single leucocyte film.

The abdominal cavity was opened up under ether-morphine narcosis. In the intervals between taking blood samples and during blood-letting the ether anesthesia was discontinued.

In our study of the hemopoietic activity of blood during the period after blood-letting, we opened up the abdominal cavity of 3 dogs 5 h after they had been bled and 3 more dogs 20 h after. In the latter 3 animals blood tests were taken once only.

The results obtained were subjected to statistical analysis. Mean data on the hemopoietic activity of the blood serum taken from the vessels under investigation was compared with data on the activity of blood from the arteries.

In addition we carried out a series of experiments designed to study the changes in hemopoietin content of the blood after a single massive blood-letting carried out after splenectomy and total gastrectomy. This series was carried out with a total of 9 dogs. We had under simultaneous observation 3 dogs: one gastrectomized, one splenectomized and a control. Fifteen blood samples were taken from each dog 48 h after it had been bled and the hemopoietic activity of each sample was determined by hemoculture using a single leucocyte film. In 3 experiments blood-letting (25 ml/kg) was carried out 5 days after operation, in 1 experiment—42 days after operation. The blood for these investigations was taken from the femoral vein.

EXPERIMENTAL RESULTS

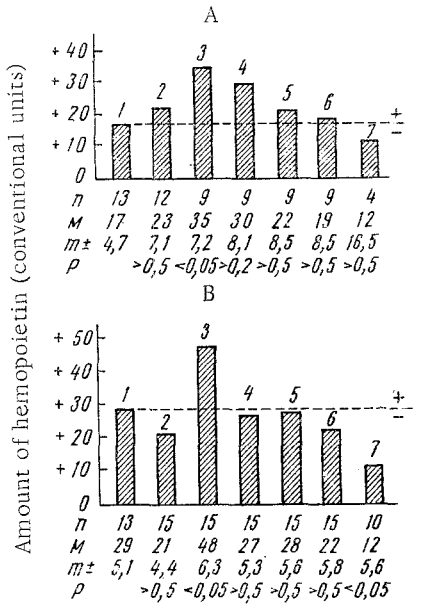

Fig. 1. Hemopoietin content of blood serum before (A) and after (B) bleeding; 1) Femoral artery; 2) femoral vein; 3) gastric vein; 4) splenic vein; 5) hepatic vein; 6) renal vein; 7) vein from small intestine.

The means for the hemopoietin content of blood serum removed from the vessels under investigation are set out in Fig. 1.

Before bleeding the mean quantity of hemopoietin per test sample taken from an artery amounted to +17. The hemopoietin content of blood from all the other vessels, except the vein from the intestine, was higher than that of the arterial blood; however, the difference was only significant in the case of the gastric vein (M+35, $P<0.05$). The hemopoietin content of blood from the splenic vein was somewhat less (M+30), the values for the hepatic vein (M+22) and the femoral (M+23) were less still. The hemopoietic activity of blood from the renal vein was only 2 units above that for arterial blood. The mean hemopoietin content of blood from the intestinal vein was 5 units below that of arterial blood.

During the first 1-2 h following bleeding the hemopoietin content of all vessels, except the vein from the small intestine, showed a considerable increase. Later on we observed periodic increases in the hemopoietic activity of blood samples from all vessels, but only the blood from the gastric vein maintained a constant high level of hemopoietin activity; this applied particularly to the last phase of observation.

After bleeding, the mean hemopoietin content of blood taken from the artery was +29. Only blood samples from the gastric vein showed a higher value (M+48, $P<0.05$) than this. Among blood samples from other vessels the mean amount of hemopoietin was less than that of arterial blood but only in the case of samples from the intestinal vein was the difference significant (M+12, $P<0.05$).

When we analyzed the data in relation to the time of sampling (Fig. 2) we found that the highest hemopoietin content was that of blood taken from the artery during the first 1-9 h after bleeding (M+39); 5-12 h after bleeding the hemopoietin content of arterial blood had fallen to +27 and by 20 h it was +29. The hemopoietin content of blood from the gastric vein was higher throughout than that of arterial blood, but the difference was only significant 20 h after bleeding (M+54, $P<0.05$). The hemopoietin content of blood from the other vessels investigated was less than that of the arterial blood; significant differences were observed in the case of the femoral vein (M+18, $P<0.05$) for blood taken during the first 1-9 h following bleeding and in the case of the intestinal vein (M+1, $P<0.05$) for blood taken during the period 5-12 h after bleeding.

Similar data from a subsequent experiment series is set out in Fig. 3. The mean amount of hemopoietin single tests of blood sera among the 3 control dogs was $+10\pm3.1$. In gastrectomized animals it was somewhat less but the difference was not statistically significant ($M\pm m = +5\pm3.4$; $P>0.5$). However, after splenectomy the hemopoietin content of the blood was significantly greater ($M\pm m = 30\pm4.1$; $P<0.001$) than among the control dogs.

The data we have obtained suggests that after blood-letting there is an emission of hemopoietin from all the organs investigated except the small intestine. Blood taken from the veins associated with these organs 1-2 h after bleeding, and also at subsequent intervals of time, possessed high hemopoietic activity. However, a more constantly high level of hemopoietin was maintained in the blood derived from the stomach, especially during the period some time after bleeding. Taking into account also the considerable reduction in hemopoietic activity of the blood following gastrectomy, as established in a previous investigation [3], we can assume that the principal source of the various hemopoietic substances is the stomach.

After splenectomy the hemopoietic activity of the blood increases, especially during the period 24-30 h after bleeding, whereas, in control animals the activity of the serum falls sharply at this time. This also is in agreement with results of our previous investigations. The high level of hemopoietins in the blood following splenectomy would appear to be related to their insufficient deposition; evidently the spleen is one of the main storage depots for hemopoietins. The possibility that the spleen manufactures hemopoietin inhibitors capable of suppressing the hemopoietic activity of the blood, should not be entirely excluded [5]. It appears to us that the low hemopoietin content of blood

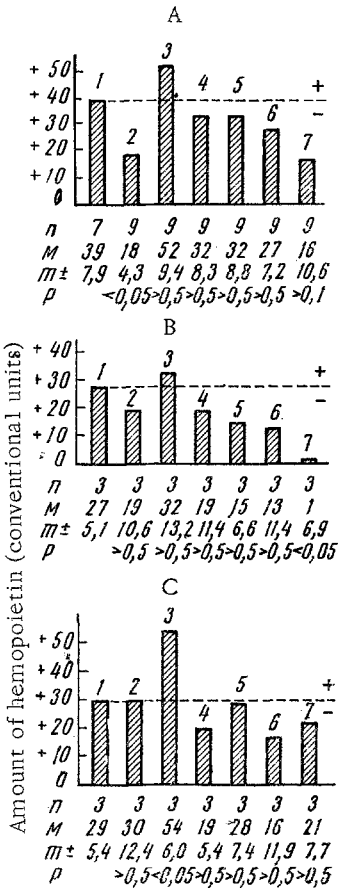

Fig. 2. Hemopoietin content of blood serum during 1st 9 h (A), 5-12 h (B), and 20 h (C) following bleeding. Symbols as in Fig. 1.

derived from the intestine could also be determined by the presence of inhibitors associated with that organ; indeed, it is possible that such substances may be associated with the intestinal flora. The significant diminution in hemopoietic activity of the blood from the femoral vein during the 1st h after bleeding may possibly be due to intense utilization of hemopoietins by the bone marrow.

There is in the literature conflicting data regarding the role of the kidneys in hemopoietin formation [4]. The results of our own investigations provide no basis for believing that the kidney is a site of hemopoietin formation. Certain blood samples taken from the renal vein did possess a high hemopoietic activity but more often blood from this source was of low activity and the mean value for hemopoietins in the renal vein was always less than that of the arterial blood.

SUMMARY

A method of hemocultures was used to study the hemopoietic activity of the serum of the blood flowing from the spleen, liver, kidney, stomach, and small intestine, as well as of that taken from the femoral artery and vein prior to and during 20 h after the stimulation by single massive bloodletting in dogs.

During the 1st hours after the blood loss the hemopoietins content increased in the blood obtained from all the vessels, excluding the intestinal vein, while a high hemopoietin level at the later periods after the bloodletting was maintained only in the blood flowing from the stomach. The average concentration of hemopoietins in the serum of the blood flowing from the gastric vein was also higher than in the arterial one. A low hemopoietin concentration was noted in the blood flowing from the intestine.

Following gastrectomy carried out 5 and 42 days before single mass blood letting, hemopoietic activity of the blood serum was reduced; it increased after splenectomy.

A conclusion was drawn that the stomach and spleen play an important role in hemopoietin regulation.

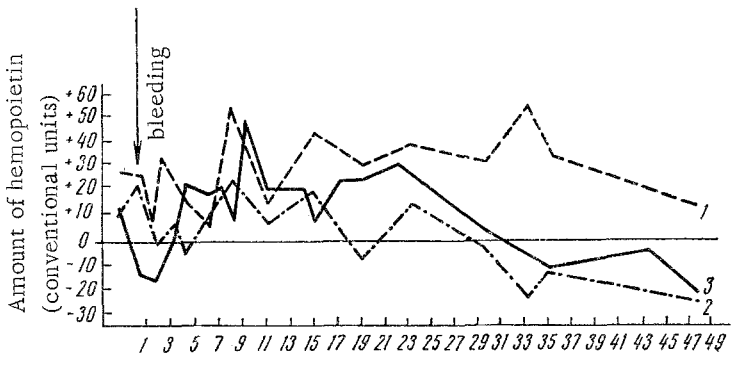

Fig. 3. Hemopoietin content of blood serum (means of 3 experiments). 1) Splenectomy; 2) gastrectomy; 3) controls.

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All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
